II

(Acts adopted under the EC Treaty/Euratom Treaty whose publication is not obligatory)

## RECOMMENDATIONS

# **COMMISSION**

#### COMMISSION RECOMMENDATION

of 18 June 2007

# on guidelines for the accommodation and care of animals used for experimental and other scientific purposes

(notified under document number C(2007) 2525)

(Text with EEA relevance)

(2007/526/EC)

THE COMMISSION OF THE EUROPEAN COMMUNITIES,

Having regard to the Treaty establishing the European Community, and in particular the second indent of Article 211 thereof,

Whereas:

- (1) Council Directive 86/609/EEC of 24 November 1986 on the approximation of laws, regulations and administrative provisions of the Member States regarding the protection of animals used for experimental and other scientific purposes (1) implements European Convention for the protection of vertebrate animals used for experimental and other scientific purposes (hereinafter 'Convention'). Council Decision 1999/575/EC (2) approved the Convention.
- (2) Under Directive 86/609/EEC Member States need to ensure that experimental animals are adequately cared for and accommodated, and that restrictions to those animals satisfying their physiological and ethological needs are only limited to the absolute minimum.

- (3) Appendix A to the Convention is implemented in Annex II to Directive 86/609/EEC, which sets out guidelines for accommodation and care of experimental animals.
- (4) On 15 June 2006 the Fourth Multilateral Consultation of Parties to the Convention adopted a revised Appendix A.
- (5) The revised Appendix A contains guidelines. Therefore it is appropriate to incorporate those guidelines in a Recommendation.
- (6) Council Directive 98/58/EC of 20 July 1998 concerning the protection of animals kept for farming purposes lays down minimum standards for the protection of animals bred or kept for farming purposes (3).
- (7) Council Regulation (EC) No 1/2005 of 22 December 2004 on the protection of animals during transport and related operations and amending Directives 64/432/EEC and 93/119/EC and Regulation (EC) No 1255/97 (4) lays down provisions on the welfare of live vertebrate animals during their transport within the Community including the

OJ L 358, 18.12.1986, p. 1. Directive as amended by Directive 2003/65/EC of the European Parliament and of the Council (OJ L 230, 16.9.2003, p. 32).

<sup>(2)</sup> OJ L 222, 24.8.1999, p. 29.

<sup>(3)</sup> OJ L 221, 8.8.1998, p. 23. Directive as amended by Regulation (EC) No 806/2003 (OJ L 122, 16.5.2003, p. 1).

<sup>(4)</sup> OJ L 3, 5.1.2005, p. 1.

specific checks to be carried out by officials on consignments entering or leaving the customs territory of the Community,

## HEREBY RECOMMENDS:

(1) Member States should pay regard to the guidelines set out in the Annex to this Recommendation for the implementation of points (a) and (b) of the first paragraph of Article 5 of Directive 86/609/EEC.

(2) Member States should inform the Commission of the steps taken to give effect to this recommendation by 15 June 2008

Done at Brussels, 18 June 2007.

For the Commission
Stavros DIMAS
Member of the Commission

# ANNEX

# Guidelines for accommodation and care of animals used for experimental and other scientific purposes

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#### INTRODUCTION

- One of the aims of Directive 86/609/EEC is to protect animals used for experimental and other scientific purposes to
  ensure that any possible pain, suffering, distress or lasting harm inflicted as a consequence of procedures being
  conducted upon them, shall be kept at a minimum.
- 2. Some procedures are conducted under field conditions on free-living, self-supporting, wild animals, but such procedures are relatively few in number. The great majority of animals used in procedures are kept in facilities ranging from outdoor corrals to cages for small animals in a laboratory animal house. This is a situation where there are often highly conflicting interests between the scientific requirements and the needs of the animal. In this conflict, the basic physiological and ethological needs of the animals (freedom of movement, social contact, meaningful activity, nutrition, water) should be restricted only for the minimum necessary period of time and degree. Such restrictions should be reviewed by scientists, animal technicians and those competent persons charged with advisory duties in relation to the well-being of the animals before procedures are undertaken to ensure that the extent of the compromise to animal welfare is minimised to a level consistent with the scientific objectives of the study.
- 3. This Annex provides guidelines for the accommodation and care of animals, based on present knowledge and good practice. It explains and supplements the basic principles adopted in Article 5 of Directive 86/609/EEC. The object of this Annex is thus to help authorities, institutions and individuals in their pursuit of the aims of Directive 86/609/EEC in this matter.
- 4. The General section provides guidelines on accommodation, housing and care relevant to all animals used for experimental and other scientific purposes. Supplementary guidance concerning commonly used species is presented in specific sections. Where no information is included in these specific sections the guidelines are included in the general section.

The species-specific sections are based on proposals made by expert groups on rodents, rabbits, dogs, cats, ferrets, non-human primates, farm species, mini-pigs, birds, amphibians, reptiles and fish. In addition to these proposals, the expert groups also submitted background information to support their proposals, based on scientific evidence and practical experiences.

This background information is the sole responsibility of the respective expert groups and is separately available. For some groups of species, namely amphibians, reptiles and fish, these explanatory documents also provide additional information on less commonly used species not referred to in the species-specific guidelines.

Should behavioural or breeding problems occur, or should further information on specific requirements for other species be required, advice should be sought from experts specialised in the species concerned and care staff, to ensure that any particular species' needs are adequately addressed.

- 5. Care is a word which, when used in connection with animals intended for, or in actual use in procedures, or in connection with laboratory animals kept for breeding purposes, covers all aspects of the relationship between animals and man. Its substance is the sum of material and non-material resources provided by man to obtain and maintain an animal in a physical and mental state where it suffers least, and promotes good science. It starts from the moment the animal is intended to be used in procedures, including breeding or keeping for that purpose, and continues until it is humanely killed or otherwise disposed of by the establishment in accordance with Article 9 of Directive 86/609/EEC after the completion of the procedure.
- 6. The Annex includes advice about the design of appropriate animal facilities and provides recommendations and guidance about how the welfare guidelines contained within Directive 86/609/EEC can be met. However, the recommended standards of space represent minimum allowances. These may have to be increased in some circumstances, as environmental requirements for individual animals might vary according, for example, to species, age, physiological conditions, stocking density and whether the animals are kept as stock, for breeding or experiments, whether long-term or short-term. Environmental enrichment is also an important factor for the welfare of the animals.
- 7. Existing facilities or equipment should be altered or replaced taking into account these guidelines, having regard to animal welfare priorities and financial and practical concerns. Pending such replacement or alteration, adjustments should be made to numbers and sizes of animals in existing enclosures in order to follow these guidelines as far as possible.

#### **DEFINITIONS**

For the purpose of these guidelines:

- (1) 'animal enclosure' means the primary accommodation in which the animals are confined, such as:
  - (a) 'cage' a permanently fixed or movable container that is enclosed by solid walls and, at least on one side, by bars or meshed wire or, where appropriate, nets, and in which one or more animals are kept or transported; depending on the stocking density and the size of the container, the freedom of movement of the animals is relatively restricted;
  - (b) 'pen' an area enclosed, for example, by walls, bars or meshed wire in which one or more animals are kept; depending on the size of the pen and the stocking density, the freedom of movement of the animals is usually less restricted than in a cage;
  - (c) 'run' an area closed, for example, by fences, walls, bars or meshed wire and frequently situated outside permanently fixed buildings, in which animals kept in cages or pens can move freely during certain periods of time in accordance with their ethological and physiological needs, such as exercise;
  - (d) 'stall' a small enclosure with three sides, usually a feed-rack and lateral separations, where one or two animals may be kept tethered;
- 2. The secondary accommodation, in which the animal enclosure(s) may be kept, are designated as 'holding rooms'.

Examples of 'holding rooms' are:

- (a) rooms where animals are normally housed, either for breeding and stocking, or during the course of a procedure;
- (b) 'containment systems', such as isolators, laminar flow cabinets and individually ventilated cage systems;

## **GENERAL SECTION**

- 1. THE PHYSICAL FACILITIES
- 1.1. Functions and general design
- 1.1.1. All facilities should be so constructed as to provide a suitable environment for the species to be kept, taking into account their physiological and ethological needs. Facilities should also be designed and managed to prevent access by unauthorised persons and the ingress or escape of animals.

Facilities that are part of a larger building complex should also be protected by appropriate security and building measures and arrangements that limit the number of entrances.

1.1.2. There should be an active maintenance programme in order to prevent and remedy any defect of buildings or equipment.

## 1.2. Holding rooms

- 1.2.1. All necessary measures should be taken to ensure regular and efficient cleaning of the rooms and the maintenance of satisfactory hygienic standards. Ceilings and walls should be damage-resistant with a smooth, impervious and easily washable surface. Special attention should be paid to junctions, including those with doors, ducts, pipes and cables. Where appropriate, an inspection window should be fitted in the door. Floors should be smooth, impervious and have a non-slippery, easily washable surface, which can carry the weight of racks and other heavy equipment without being damaged. Drains, if present, should be adequately covered and fitted with a barrier, which will prevent vermin from gaining access or animals from escaping.
- 1.2.2. Where the animals are allowed to run freely, walls and floors should be surfaced with a material resistant to the heavy wear and tear caused by the animals and the cleaning process. The material should not be detrimental to the health of the animals and should be such that the animals cannot hurt themselves. Additional protection should be given to any equipment or fixtures so that they may not be damaged by the animals or injure the animals themselves.

- 1.2.3. Species that are incompatible, for example predator and prey, or animals requiring different environmental conditions, should not be housed in the same room nor, in the case of predator and prey, within sight, smell or sound.
- 1.2.4. Holding rooms should, where appropriate, be provided with facilities for carrying out minor procedures and manipulations.

#### 1.3. General and special purpose procedure rooms

- 1.3.1 At breeding or supplying establishments suitable facilities for making consignments of animals ready for dispatch should be available.
- 1.3.2. All establishments should also have available, as a minimum, laboratory facilities for the carrying out of simple diagnostic tests, post-mortem examinations, and/or the collection of samples which are to be subjected to more extensive laboratory investigations elsewhere.
- 1.3.3. Facilities should be provided to enable newly-acquired animals to be isolated until their health status can be determined, and the potential health risk to established animals assessed and minimised.
- 1.3.4. General and special purpose procedure rooms should be made available for situations where it is undesirable to carry out the procedures or observations in the holding rooms.
- 1.3.5. Where appropriate, there should be provision for one or more separate rooms suitably equipped for the performance of surgical procedures under aseptic conditions. There should be facilities for post-operative recovery where this is warranted.
- 1.3.6. There should be accommodation for separate housing of sick or injured animals, where necessary.

#### 1.4. Service rooms

- 1.4.1. Storerooms should be designed, used and maintained to safeguard the quality of food and bedding. These rooms should be vermin and insect-proof. Other materials, which may be contaminated or present a hazard to animals or staff, should be stored separately.
- 1.4.2. Separate storerooms for clean cages, instruments and equipment should be provided.
- 1.4.3. The cleaning and washing areas should be large enough to accommodate the installations necessary to decontaminate and clean used equipment. The cleaning process should be arranged so as to separate the flow of clean and dirty equipment to prevent the contamination of newly-cleaned equipment. Walls and floors should be covered with a suitably durable surface material and the ventilation system should have ample capacity to carry away the excess heat and humidity.
- 1.4.4. Provision should be made for the hygienic storage and disposal of carcasses and animal waste. If incineration on the site is not possible or necessary, suitable arrangements should be made for the safe disposal of such material, having regard to national and local regulation and by-laws. Special precautions should be taken with toxic, radioactive or infectious waste.
- 1.4.5. The general design and construction of circulation areas should correspond to the standards of the holding rooms. The corridors should be wide enough to allow easy circulation of movable equipment.
- 2. THE ENVIRONMENT AND ITS CONTROL

#### 2.1. Ventilation

- 2.1.1. Adequate ventilation should be provided in the holding room and the animal enclosures to satisfy the requirements of the animals housed. The purpose of the ventilation system is to provide sufficient fresh air of an appropriate quality and to keep down the levels and spread of odours, noxious gases, dust and infectious agents of any kind. It also provides for the removal of excess heat and humidity.
- 2.1.2. The air in the room should be renewed at frequent intervals. A ventilation rate of fifteen to twenty air changes per hour is normally adequate. However, in some circumstances, for example where stocking density is low, eight to ten air changes per hour may suffice. In some cases, natural ventilation may suffice and mechanical ventilation

may not even be needed. Recirculation of untreated air should be avoided. However, it should be emphasised that even the most efficient system cannot compensate for poor cleaning routines or negligence.

- 2.1.3. The ventilation system should be so designed as to avoid harmful draughts and noise disturbance.
- 2.1.4. Smoking in rooms where there are animals should be forbidden.

#### 2.2. **Temperature**

2.2.1. The subsequent species-specific sections give the range within which it is recommended that the temperature should be maintained. It should also be emphasised that the figures given in these sections apply only to adult, normal animals. New-born, young, hairless, newly-operated, sick or injured animals will often require a much higher temperature level. The temperature of the premises should be regulated according to possible changes in the animals' thermal regulation, which may be compromised due to special physiological conditions or to the effects of the procedures.

Temperature in the holding rooms should be measured and logged on a daily basis.

- 2.2.2. It may be necessary to provide a ventilation system having the capacity both to heat and cool the air supplied.
- 2.2.3. In user establishments a precise temperature control in the holding rooms may be required, because the temperature of the environment is a physical factor which has a profound effect on the metabolism and behaviour of all animals, and therefore affects the validity of certain scientific outcomes.
- 2.2.4. Outdoor areas provided for animals to exercise and interact cannot have strict temperature regulation. Animals should not be restricted to such areas under climatic conditions which may cause them distress.

# 2.3. Humidity

For some species, such as rats and gerbils, the relative humidity may need to be controlled within a fairly narrow range to minimise the possibility of health or welfare problems, whereas other species, such as dogs, tolerate well wide fluctuations in humidity levels.

## 2.4. Lighting

Where natural light does not provide an appropriate light/dark cycle, it is necessary to provide controlled lighting both to satisfy the biological requirements of the animals and to provide a satisfactory working environment. Exposure of some species to bright light should be avoided and darker areas for withdrawal should be available within the animal enclosures. There should be adequate illumination for the performance of husbandry procedures and inspection of the animals. Regular photoperiods and intensity of light suitable to the species should be provided and interruptions to these should be avoided. When keeping albino animals, one should take into account their sensitivity to light. Consideration should be given to the inclusion of windows in holding rooms, since they are a source of natural light and can provide environmental enrichment for some species, especially non-human primates, dogs, cats, some farm animals and other large mammals.

#### 2.5. Noise

Noise can be a disturbing factor for animals. High noise levels and sudden noises can cause stress which, in addition to the welfare consequences for the animal, may influence experimental data. Noise levels within the hearing ranges of animals, including in some cases ultrasound, that is, sound above the hearing range of the human being, conventionally taken to be sounds exceeding 20 kHz, should be minimised particularly during their resting phase. Alarm systems should sound outside the sensitive hearing range of the animals, where this does not conflict with their audibility to humans. The layout of rooms and corridors can be major factors influencing the acoustic environment and this should be taken into account in their design. Holding rooms should be provided with adequate noise insulation and absorption materials.

## 2.6. Alarm systems

A technologically dependent animal facility is a vulnerable entity. It is strongly recommended that such facilities are appropriately protected to detect hazards such as fires, the intrusion of unauthorised persons, and the breakdown of essential equipment, such as ventilation fans, air heaters or coolers and humidifiers.

Animal facilities which rely heavily on electrical or mechanical equipment for environmental control and protection should have a stand-by system to maintain essential services and emergency lighting systems as well as to ensure that alarm systems themselves do not fail to operate.

Heating and ventilation systems should be equipped with monitoring devices and alarms to ensure that any faults can be quickly identified and promptly rectified.

Clear instructions on emergency procedures should be prominently displayed. Alarms are recommended for water tanks for fish and other aquatic animals in case of failure of the water or air supply. Care should be taken to ensure that the operation of an alarm system causes as little disturbance as possible to the animals.

#### 3. EDUCATION AND TRAINING

All persons involved in caring for, or otherwise involved with, animals being bred, held or used for experimental or other scientific purposes should be appropriately educated and trained to the standard recommended in the Resolution on education and training of persons working with laboratory animals adopted by the Multilateral Consultation of the Parties to the Council of Europe Convention ETS No 123 on 3 December 1993.

#### 4. CARE

#### 4.1. Health

4.1.1. Animals within an animal facility are totally dependent on humans for their health and well-being. The physical and psychological state of the animals will be influenced by their local environment, food, water and the care and attention provided by the animal care staff.

A strategy should be in place in all establishments to ensure that an appropriate health status is maintained, which safeguards animal welfare and meets scientific requirements. This strategy should include a microbiological surveillance programme, plans for dealing with health breakdowns, and should define health parameters and procedures for the introduction of new animals.

- 4.1.2. The person responsible for the establishment should ensure regular inspection of the animals and supervision of the accommodation and care by a veterinarian or other competent person. Inspection of the animals should be made at least daily by a person trained in accordance with paragraph 3 of the General section, to ensure that all sick or injured animals are identified and appropriate action taken. Regular health monitoring should be carried out
- 4.1.3. Because of the potential risk of contamination of animals and staff presented by the handling of animals, particular attention should be paid to the institution of hygiene procedures and supervision of staff health.

## 4.2. Capture from the wild

- 4.2.1. When animals need to be captured, it should only be done by humane methods and by persons competent to apply them. The impact of the capturing procedures on the remaining wildlife and habitats should be minimised.
- 4.2.2. Any animal found, at or after capture, to be injured or in poor health should be examined by a competent person as soon as possible, and appropriate action taken. This may require referral to a veterinarian for treatment, or, in the case of serious injury, the animal should be killed immediately by a humane method, in line with the principles set out in the European Commission Recommendations for the euthanasia of experimental animals (Part 1 and Part 2). Appropriate and sufficient transport containers and means of transport should be available at capture sites, in case animals need to be moved for examination or treatment.

4.2.3. Special consideration should be given to the acclimatisation, quarantine, housing, husbandry and care of wild caught animals. The eventual fate of wild caught animals following the conclusion of scientific procedures should also be given due consideration before the work begins. This is to ensure that the practical difficulties and welfare issues associated with any subsequent release to the wild can be satisfactorily addressed.

#### 4.3. Transport of animals

4.3.1. For animals, transportation is a stressful experience which should be mitigated as far as possible. The following principles should apply to all animal movements, from short journeys by vehicle within scientific establishments to international transportation.

When Council Regulation (EC) 1/2005 ( $^1$ ) is applied, regard should be paid to the Resolution on the acquisition and transport of laboratory animals, adopted by the May 1997 Multilateral Consultation of the Parties to Council of Europe Convention (ETS No 123).

- 4.3.2. Both sender and recipient should agree the conditions of transport, departure and arrival times to ensure that full preparation can be made for the animals' arrival. The sender should ensure that the animals are examined and found to be fit for transport before being placed in the transport container.
- 4.3.3. Animals that are sick or injured should not be considered fit for transport, except for slightly injured or sick animals whose transport would not cause additional suffering, or where the transport is under veterinary supervision for, or following, veterinary treatment.

Sick or injured animals may also be transported for experimental or other scientific purposes approved by the relevant competent authority, if the illness or injury is part of the research programme. No additional suffering should be imposed by the transport of such animals, and particular attention should be paid to any additional care which may be required. A competent person should confirm that such animals are fit for the intended journey.

- 4.3.4. The person responsible for the transport of the animals has the overall control over the organisation, carrying out and completion of the whole journey, regardless of whether duties are subcontracted to other Parties during transport.
- 4.3.5. The person in charge of the welfare of the animals has direct physical responsibility for the care of the animals during transport. Such a person may be the attendant or the driver of a vehicle if fulfilling the same role. The person in charge of the welfare of animals being transported should be aware of the special needs of the laboratory animals in their care.
- 4.3.6. The route should be planned in order to ensure that the transport is carried out efficiently to minimise journey time, from loading to unloading, and to avoid delays in order to limit any stress and suffering of the animals. Care is needed to ensure that animals are maintained under suitable environmental conditions for the species, and that measures are taken to minimise sudden movements, excessive noise, or vibration during transport.
- 4.3.7. Where appropriate, the container should be designed to prevent or restrict the entry or spread of microorganisms. It should allow visual inspection of the animals without compromising the microbiological status of the animals.
- 4.3.8. On arrival at their destination the animals should be removed from their transport containers and examined by a competent person with the least possible delay. Animals, which are sick, injured or otherwise out of condition, should be kept under close observation and housed separately from other animals. These animals should be provided with veterinary treatment as appropriate or, if deemed necessary, promptly killed by a humane method.

## 4.4. Quarantine, acclimatisation and isolation

The objectives of quarantine and isolation periods are:

- a. to protect other animals in the establishment;
- b. to protect man against zoonotic infection; and

c. together with an acclimatisation period, to foster good scientific practice.

According to the circumstances, these periods may vary and are either determined by the national regulations of the Member State, or a competent person normally the veterinarian appointed by the establishment.

#### Quarantine

Quarantine is considered as a period of housing newly introduced or reintroduced animals separate from existing animals in the establishment to establish the state of health of the animals and to prevent the introduction of disease. Such a period is recommended when the health status of the animal is not known.

#### Acclimatisation

A period of acclimatisation is needed to allow animals to recover from transport stress, to become accustomed to a new environment and to husbandry and care practices. Even when the animals are seen to be in good health, it is necessary for them to undergo a period of acclimatisation before being used in a procedure. The time required depends on several factors, such as the stress to which the animals have been subjected which in turn depends on several factors such as the duration of the transportation and the age of the animal and change of the social environment. It should also be taken into account that international transport may necessitate an extended period of acclimatisation due to disturbance of the diurnal rhythm of the animals.

#### Isolation

A period of isolation is intended to reduce the risk of infection to other animals or humans. Any animal suspected of posing such a risk should be housed in a separate facility.

## 4.5. Housing and enrichment

## 4.5.1. Introduction

All animals should be allowed adequate space to express a wide behavioural repertoire. Animals should be socially housed wherever possible and provided with an adequately complex environment within the animal enclosure to enable them to carry out a range of normal behaviours. Restricted environments can lead to behavioural and physiological abnormalities and affect the validity of scientific data.

Consideration should be given to the potential impact of the type of accommodation, and that of the environmental and social enrichment programmes, on the outcome of scientific studies, in order to avoid the generation of invalid scientific data and consequential animal wastage.

The housing and enrichment strategies used in breeding, supplying and user establishments should be designed to fulfil the needs of the species housed and to ensure that the animals can make the best use of the space available. Their design should also take into account the need to observe the animals with minimum disruption and to facilitate handling. Suggested minimum animal enclosure sizes and space allowances are included in the subsequent individual species sections.

Unless otherwise specified, additional surface areas provided by enclosure additions, such as shelves, should be provided in addition to the recommended minimum floor areas.

# 4.5.2. Housing

Animals, except those which are naturally solitary, should be socially housed in stable groups of compatible individuals. Single housing should only occur if there is justification on veterinary or welfare grounds. Single housing on experimental grounds should be determined in consultation with the animal technician and with the competent person charged with advisory duties in relation to the well-being of the animals. In such circumstances, additional resources should be targeted to the welfare and care of these animals. In such cases, the duration should be limited to the minimum period necessary and, where possible, visual, auditory, olfactory and tactile contact should be maintained. The introduction or re-introduction of animals to established groups should be carefully monitored by adequately trained staff, to avoid problems of incompatibility and disrupted social relationships. The possibility of social housing should be promoted by purchasing compatible individuals when procuring animals of gregarious species.

#### 4.5.3. Enrichment

All animals should be provided with sufficient space of adequate complexity to allow expression of a wide range of normal behaviour. They should be given a degree of control and choice over their environment to reduce stress-induced behaviour. This may be achieved by using appropriate enrichment techniques, which extend the range of activities available to the animal and increase their coping activities. In addition to social activities, enrichment can be achieved by allowing and promoting physical exercise, foraging, manipulative and cognitive activities, as appropriate to the species. It is advisable to allow the animals to exercise at every possible opportunity. Environmental enrichment in animal enclosures should be appropriate to the species-specific and individual needs of the animals concerned. Forms of enrichment should be adaptable so that innovation based on new understanding may be incorporated. The enrichment programme should be regularly reviewed and updated. The staff responsible for animal care should understand the natural behaviour and biology of the species, so that they can make sensible and informed choices on enrichment. They should be aware that all enrichment initiatives are not necessarily to the advantage of the animal and therefore should monitor their effects and adjust the programme as required.

#### 4.5.4. Animal enclosures

Animal enclosures should not be made out of materials detrimental to the health of the animals. Their design and construction should be such that no injury to the animals is caused. Unless they are disposable, they should be made from materials that will withstand cleaning and decontamination techniques. In particular, attention should be given to the design of animal enclosure floors, which should be appropriate to the species and age of the animals and be designed to facilitate the removal of excreta.

#### 4.6. Feeding

- 4.6.1. The form, content and presentation of the diet should meet the nutritional and behavioural needs of the animal. For some species, the opportunity for foraging should be given. Roughage is an important component of the diet for some species of animals, as well as a means of satisfying some behavioural needs.
- 4.6.2. The animals' diet should be palatable and non-contaminated. In the selection of raw materials, production, preparation and presentation of feed, precautions should be taken to minimise chemical, physical and microbiological contamination. The feed should be packed in bags that provide clear information on the identity of the product and its date of production. An expiry date should be clearly defined by the manufacturer and adhered to.

Packing, transport and storage should also be such as to avoid contamination, deterioration or destruction. Storerooms should be cool, dark, dry and vermin- and insect-proof. Perishable feed like greens, vegetables, fruit, meat, fish should be stored in cold rooms, refrigerators or freezers.

All feed hoppers, troughs or other utensils used for feeding should be regularly cleaned and, if necessary, sterilised. If moist feed is used, or if the feed is easily contaminated with for example water or urine, daily cleaning is necessary.

4.6.3. Each animal should be able to access the food, with sufficient feeding space provided to limit competition. In some circumstances, food intake may need to be controlled to avoid obesity.

## 4.7. Watering

- 4.7.1. Uncontaminated drinking water should always be available to all animals. Water is, however, a vehicle for microorganisms, and the supply should therefore be so arranged that the contamination risk is minimised.
- 4.7.2. Watering systems should be designed and used to provide an adequate quantity of water of suitable quality. Sufficient watering points (drinkers) should be available. When automatic watering systems are used, their functioning should be regularly checked, serviced and flushed to avoid accidents, such as blockages or leakages and the spread of infections. If solid-bottomed cages are used, care should be taken to minimise the risk of flooding.
- 4.7.3. In fishes, amphibians and reptiles, tolerance for acidity, chlorine and many other chemicals differs widely from species to species. Therefore provision should be made to adapt the water supply for aquaria and tanks to the needs and tolerance limits of the individual species.

## 4.8. Flooring, substrate, litter, bedding and nesting material

4.8.1. Appropriate bedding materials or sleeping structures should always be provided for animals, as well as appropriate nesting materials or structures for breeding animals.

Various materials are commonly placed into the animal enclosure to serve the following functions: to absorb urine and faeces, and thus facilitate cleaning: to allow the animal to perform certain species-specific behaviour, such as foraging, digging or burrowing; to provide a comfortable, yielding surface or secure area for sleeping; to allow the animal to build a nest for breeding purposes.

Certain materials may not serve all of these needs, and it is therefore important to provide sufficient and appropriate materials. Any such materials should be dry, absorbent, dust-free, non-toxic and free from infectious agents or vermin and other forms of contamination. Materials derived from wood that has been chemically treated or containing toxic natural substances as well as products which cannot be clearly defined and standardised should be avoided.

4.8.2. Within the animal enclosure, the flooring should provide a solid, comfortable resting area for all animals. All sleeping areas should be kept clean and dry.

## 4.9. Cleaning

- 4.9.1. The standard of a facility, including good husbandry, depends very much on good hygiene. A very high standard of cleanliness and order should also be maintained in holding, washing and storage rooms. Adequate routines for the cleaning, washing, decontamination and, when necessary, sterilisation of enclosures and accessories, bottles and other equipment should be established and carried out.
- 4.9.2. These cleaning and disinfection regimes should not be detrimental to animal health or welfare. Clear operating procedures, including a recording system, should be in place for the changing of bedding in animal enclosures.
- 4.9.3. There should be regular cleaning and, where appropriate, renewal of the materials forming the ground surface in animal enclosures to avoid them becoming a source of infection and parasite infestation.
- 4.9.4. Odour-marking is an important form of behaviour in some species, and cleaning disturbances will cause some degree of social disruption. Cleaning regimes should have regard for these behavioural needs. Decisions on frequency of cleaning should be based on the type of animal enclosure, the type of animal, the stocking density, and the ability of the ventilation system to maintain suitable air quality.

#### 4.10. Handling

The quality of care animals are given in the laboratory may influence not only breeding success, growth rate and welfare, but also the quality and outcome of experimental procedures. Accustoming animals to competent and confident handling during routine husbandry and procedures reduces stress both to animals and personnel. For some species, for example dogs and non-human primates, a training programme to encourage cooperation during procedures can be beneficial to the animals, the animal care staff and the scientific programme. For certain species, social contact with humans should be a priority.

However, in some cases, handling should be avoided. This may be particularly the case with wild animals, and is one reason why wild animals can be less suitable as experimental subjects. Staff caring for animals are expected, at all times, to have a caring and respectful attitude towards the animals in their care, and to be proficient in the handling and restraint of the animals.

Where appropriate, staff time should be set aside for talking to, handling, training and grooming animals.

## 4.11. Humane killing

4.11.1. All humane methods of killing animals require expertise, which can only be attained by appropriate training. Animals should be killed using a method that adheres to the principles set by the European Commission Recommendations for the euthanasia of experimental animals (Part 1 and Part 2).

4.11.2. A deeply unconscious animal can be exsanguinated, but drugs which paralyse muscles before unconsciousness occurs, drugs with curariform effects and electrocution without passage of current through the brain, should not be used without prior anaesthesia.

Disposal should not be allowed until death has been confirmed.

#### 4.12. Records

Records of source, use and final disposal of all animals bred, kept for breeding, or for subsequent supply for use in scientific procedures should be used not only for statistical purposes but, in conjunction with health and breeding records, as indicators of animal welfare and for husbandry and planning purposes.

#### 4.13. **Identification**

In some instances, it is necessary for animals to be individually identified, for example, when being used for breeding purposes or scientific procedures, to enable accurate records to be kept. The method chosen should be reliable and cause the minimum pain and discomfort to the animal when applied and in the long-term. Sedatives or local anaesthetics and analgesics should be used if necessary. Staff should be trained in carrying out the identification and marking techniques.

#### SPECIES-SPECIFIC SECTION

#### A. SPECIES-SPECIFIC GUIDELINES FOR RODENTS

#### 1. **Introduction**

Mice

The laboratory mouse is derived from the wild house mouse (*Mus musculus*) a largely nocturnal burrowing and climbing animal which builds nests for regulation of the microenvironment, shelter and reproduction. Mice are good climbers. Mice do not readily cross open spaces, preferring to remain close to walls or other structures. A wide range of social organisations has been observed depending on population density and intense territoriality may be seen in reproductively active males. Pregnant and lactating females may prove aggressive in nest defence. As mice, particularly albino strains, have poor sight they rely heavily on their sense of smell and create patterns of urine markings in their environment. Mice also have very acute hearing and are sensitive to ultrasound. There are considerable differences in the expression and intensity of behaviour depending on the strain.

Rats

The laboratory rat is derived from the wild brown rat (*Rattus norvegicus*) and is a highly social animal. Rats avoid open spaces, and use urine to mark territory. Their sense of smell and hearing are highly developed, and rats are particularly sensitive to ultrasound. Daylight vision is poor, but dim-light vision is effective in some pigmented strains. Albino rats avoid areas with light levels over 25 lux. Activity is greater during hours of darkness. Young animals are very exploratory and often engage in social play.

## Gerbils

The gerbil or Mongolian jird (*Meriones sp.*) is a social animal and is largely nocturnal, although in the laboratory it is also active during daylight. In the wild, gerbils build burrows with tunnel entrances as a protection against predators, and in the laboratory often develop stereotypic digging behaviour unless provided with adequate facilities.

#### Hamsters

The wild ancestors (*Mesocricetus sp.*) of the laboratory hamster are largely solitary. The female hamster is larger and more aggressive than the male and can inflict serious injury on her mate. Hamsters often make a latrine area within the enclosure, mark areas with secretions from a flank gland, and females frequently selectively reduce the size of their own litter by cannibalism.

#### Guinea Pigs

Wild guinea pigs (*Cavia porcellus*) are social, cursorial rodents which do not burrow, but live under cover and may use burrows made by other animals. Adult males may be aggressive to each other, but generally aggression is rare. Guinea pigs tend to freeze at unexpected sounds and may stampede as a group in response to sudden unexpected movements. Guinea pigs are extremely sensitive to being moved and may freeze as a result for thirty minutes or more.

#### 2. The environment and its control

#### 2.1. Ventilation

(See paragraph 2.1. of the General section)

## 2.2. Temperature

Rodents should be maintained within a temperature range of 20  $^{\circ}$ C to 24  $^{\circ}$ C. Local temperatures among groups of rodents in solid-floored enclosures will often be higher than room temperatures. Even with adequate ventilation the enclosure temperatures may be up to 6  $^{\circ}$ C above room temperature. Nesting material/nestboxes give animals the opportunity to control their own microclimate. Special attention should be paid to the temperature in containment systems as well as to that provided for hairless animals.

## 2.3. Humidity

The relative humidity in rodent facilities should be kept at 45 to 65 %. Excepted from this principle are gerbils, which should be kept at a relative humidity of 35 to 55 %.

#### 2.4. Lighting

Light levels within the enclosure should be low. All racks should have shaded tops to reduce the risk of retinal degeneration. This is of particular importance for albino animals.

A period of red light at frequencies undetectable to the rodents can be useful during the dark period so that staff can monitor the rodents in their active phase.

## 2.5. Noise

As rodents are very sensitive to ultrasound, and use it for communication, it is important that this extraneous noise is minimised. Ultrasonic noise (over 20 kHz) produced by many common laboratory fittings, including dripping taps, trolley wheels and computer monitors, can cause abnormal behaviour and breeding cycles. It may be advisable to monitor the acoustic environment over a broad range of frequencies and over extended time periods.

## 2.6. Alarm systems

(See paragraph 2.6. of the General section)

## 3. Health

(See paragraphs 4.1. and 4.4. of the General section)

# 4. Housing, enrichment and care

# 4.1. Housing

Gregarious species should be group-housed as long as the groups are stable and harmonious. Such groups can be achieved, although it is difficult, when housing male mice, adult hamsters or gerbils, as this can result in severe conspecific aggression.

Animals may be housed individually if adverse effects or damage are likely to occur. Disruption of established stable and harmonious groups should be minimised, as this can be very stressful.

#### 4.2. Enrichment

The enclosures and their enrichment should allow the animals to manifest normal behaviours and to enable conspecifics to reduce competitive situations adequately.

Bedding and nesting material and refuges are very important resources for rodents in breeding, stock or under procedure and should be provided unless there is a justification on veterinary or welfare grounds against doing so. Withholding of such materials on experimental grounds should be agreed with the animal technician and with the competent person charged with advisory duties in relation to the well-being of the animals. Nesting materials should allow the rodents to manipulate the material and construct a nest. Nest boxes should be provided if insufficient nesting material is provided for the animals to build a complete, covered nest. Bedding materials should absorb urine and may be used by the rodents to lay down urine marks. Nesting material is important for rats, mice, hamsters and gerbils as it enables them to create appropriate microenvironments for resting and breeding. Nest boxes or other refuges are important for guinea pigs, hamsters and rats.

Guinea pigs should always be provided with manipulable materials such as hay for chewing and concealment.

Wood sticks for chewing and gnawing may be considered for enrichment for all rodent species.

Many rodent species attempt to divide up their own enclosures into areas for feeding, resting, urination and food storage. These divisions may be based on odour marks rather than physical division but partial barriers may be beneficial to allow the animals to initiate or avoid contact with other group members. To increase environmental complexity the addition of some form of enclosure enrichment is strongly recommended. Tubes, boxes and climbing racks are examples of devices which have been used successfully for rodents, and these can have the added benefit of increasing utilisable floor area.

Gerbils need comparatively more space than other rodent species in order to allow them to build and/or use burrows of sufficient size. Gerbils require a thick layer of litter for digging and nesting or a burrow substitute, which needs to be at least 20 cm long.

Consideration should be given to the use of translucent or tinted enclosures and inserts which permit good observation of the animals without disturbing them.

The same principles regarding quality and quantity of space, environmental enrichment and other considerations in this document should apply to containment systems such as individually ventilated cages (IVCs), although the design of the system may mean that these may have to be approached differently.

#### 4.3. Enclosures — dimensions and flooring

The enclosures should be made of easy-to-clean materials and their design should allow proper inspection of the animals without causing disturbance to them.

Once young animals become active they require proportionally more space than adults do.

## 4.3.1. Dimensions

In this and subsequent tables for all rodent recommendations 'enclosure height' means the vertical distance between the enclosure floor and the top of the enclosure, and this height should apply over more than  $50\,\%$  of the minimum enclosure floor area prior to the addition of enrichment devices.

When designing procedures, consideration should be given to the potential growth of the animals to ensure adequate space is provided (as detailed in Tables A.1. to A.5) for the duration of the study.

Table A.1.

Mice: Minimum enclosure dimensions and space allowances

	Body weight (g)	Minimum enclosure size (cm²)	Floor area per animal (cm²)	Minimum enclosure height (cm)
In stock and during pro- cedures	up to 20	330	60	12
cedures	over 20 to 25	330	70	12
	over 25 to 30	330	80	12
	over 30	330	100	12
Breeding		330		
		For a monogamous pair (outbred/inbred) or a trio (inbred). For each additional female plus litter 180 cm <sup>2</sup> should be		
		added.		12
Stock at breeders (*) Enclosure size	less than 20			
950 cm <sup>2</sup>		950	40	12
Enclosure size 1 500 cm <sup>2</sup>	less than 20	1 500	30	12

<sup>(\*)</sup> Post-weaned mice may be kept at these higher stocking densities, for the short period after weaning until issue, provided that the animals are housed in larger enclosures with adequate enrichment. These housing conditions should not cause any welfare deficit such as: increased levels of aggression, morbidity or mortality, stereotypes and other behavioural deficits, weight loss, or other physiological or behavioural stress responses.

Table A.2.

Rats: Minimum enclosure dimensions and space allowances

	Body weight (g)	Minimum enclosure size (cm²)	Floor area per animal (cm²)	Minimum enclosure height (cm)
In stock and during pro-	up to 200	800	200	18
cedures (*)	over 200 to 300	800	250	18
	over 300 to 400	800	350	18
	over 400 to 600	800	450	18
	over 600	1 500	600	18
Breeding		800		
		Mother and litter. For each additional adult animal permanently		
		added to the enclosure add 400 cm <sup>2</sup>		18
Stock at breeders (**)	up to 50	1 500	100	18
	over 50 to 100	1 500	125	18
Enclosure size	over 100 to 150	1 500	150	18
1 500 cm <sup>2</sup>	over 150 to 200	1 500	175	18

	Body weight (g)	Minimum enclosure size (cm²)	Floor area per animal (cm²)	Minimum enclosure height (cm)
Stock at breeders (**)	up to 100	2 500	100	18
	over 100 to 150	2 500	125	18
Enclosure size 2 500 cm <sup>2</sup>	over 150 to 200	2 500	150	18

Table A.3. Gerbils: Minimum enclosure dimensions and space allowances

	Body weight (g)	Minimum enclosure size (cm²)	Floor area per animal (cm²)	Minimum enclosure height (cm)
In stock and during procedures	up to 40	1 200	150	18
	over 40	1 200	250	18
Breeding		1 200		18
		Monogamous pair or trio with offspring		

Table A.4. Hamsters: Minimum enclosure dimensions and space allowances

	Body weight (g)	Minimum enclosure size (cm²)	Floor area per animal (cm²)	Minimum enclosure height (cm)
In stock and during procedures	up to 60	800	150	14
	over 60 to 100	800	200	14
	over 100	800	250	14
Breeding		800		
		Mother or monogamous pair with litter		14
Stock at breeders (*)	less than 60	1 500	100	14

Post-weaned hamsters may be kept at these stocking densities, for the short period after weaning until issue, provided that the animals are housed in larger enclosures with adequate enrichment. These housing conditions should not cause any welfare deficit such as: increased levels of aggression, morbidity or mortality, stereotypies and other behavioural deficits, weight loss, or other physiological or behavioural stress responses.

 <sup>(\*)</sup> In lifetime studies, animals should be provided with enclosures of a suitable size to enable the animals to be socially housed. As stocking densities towards the end of such studies may be difficult to predict, consequentially there may be occasions where space allowances per individual animal may fall below those indicated above. In such circumstances priority should be given to maintaining stable social structures.
 (\*\*) Post-weaned rats may be kept at these stocking densities, for the short period after weaning until issue, provided that the animals are housed in larger enclosures with adequate enrichment. These housing conditions should not cause any welfare deficit such as: increased levels of aggression, morbidity or mortality, stereotypies and other behavioural deficits, weight loss, or other physiological or behavioural stress responses.

Table A.5.

Guinea pigs: Minimum enclosure dimensions and space allowances

	Body weight (g)	Minimum enclosure size (cm²)	Floor area per animal (cm²)	Minimum enclosure height (cm)
In stock and during procedures	up to 200 over 200 to 300 over 300 to 450 over 450 to 700 over 700	1 800 1 800 1 800 2 500 2 500	200 350 500 700 900	23 23 23 23 23 23
Breeding		2 500 Pair with litter. For each additional breeding female add 1 000 cm <sup>2</sup>		23

#### 4.3.2. Flooring

Solid floors with bedding or perforated floors are preferable to grid or wire mesh floors. If grids or wire mesh are used, a solid or bedded area or, as an alternative in the case of guinea pigs, a slatted area, should be provided for the animals to rest on unless specific experimental conditions prevent this. Bedding may be withheld as part of time-mating practices.

As mesh floors can lead to serious injuries, the floors should be closely inspected and maintained to ensure that there are no loose or sharp projections.

During late pregnancy, parturition and lactation, breeding females should only be kept on solid floors with bedding.

#### 4.4. Feeding

(See paragraph 4.6. of the General section)

## 4.5. Watering

(See paragraph 4.7. of the General section)

## 4.6. Substrate, litter, bedding and nesting material

(See paragraph 4.8. of the General section)

## 4.7. Cleaning

Although high hygiene standards should be maintained, it may be advisable to maintain some odour cues left by animals. Too frequent changing of enclosures should be avoided, particularly where pregnant animals and females with litters are concerned, as such disturbances can result in mis-mothering or cannibalism.

Decisions on frequency of cleaning should therefore be based on the type of the enclosure, type of animal, stocking densities, and the ability of ventilation systems to maintain suitable air quality.

## 4.8. Handling

When handling, care needs to be taken to minimise disturbance of the animals or their enclosure environment. This is of particular importance with hamsters.

## 4.9. Humane killing

(See paragraph 4.11. of the General section)

#### 4.10. Records

(See paragraph 4.12. of the General section)

#### 4.11. Identification

(See paragraph 4.13. of the General section)

#### B. SPECIES-SPECIFIC GUIDELINES FOR RABBITS

#### 1. **Introduction**

The rabbit (Oryctolagus cuniculi) is a naturally gregarious species. Rabbits should be allowed adequate space and an enriched environment, the denial of which can result in loss of normal locomotor activity and in skeletal abnormalities.

#### 2. The environment and its control

#### 2.1. Ventilation

(See paragraph 2.1. of the General section)

# 2.2. Temperature

Rabbits should be maintained in a temperature range of  $15\,^{\circ}$ C to  $21\,^{\circ}$ C. Local temperatures among groups of rabbits in solid-floored enclosures will often be higher than room temperatures. Even with adequate ventilation the enclosure temperatures may be up to  $6\,^{\circ}$ C above room temperature.

Nesting material/nestboxes give animals the opportunity to control their own microclimate. Special attention should be paid to the temperature in containment systems.

## 2.3. Humidity

The relative humidity in rabbit facilities should not be less than 45 %.

## 2.4. Lighting

(See paragraph 2.4. of the General section)

#### 2.5. Noise

(See paragraph 2.5. of the General section)

## 2.6. Alarm systems

(See paragraph 2.6. of the General section)

#### 3. Health

(See paragraphs 4.1. and 4.4. of the General section)

## 4. Housing, enrichment and care

## 4.1. Housing

Young and female rabbits should be housed in harmonious social groups. Single housing should only occur if there is justification on veterinary or welfare grounds. Single housing on experimental grounds should be determined in consultation with the animal technician and with the competent person charged with advisory duties in relation to the well-being of the animals. Adult entire males may perform territorial behaviour and should not be housed with other entire males. Enriched floor pens have been used with success to house young rabbits and adult female rabbits although groups may need to be carefully managed to avoid aggression. Ideally rabbits for group housing should be littermates that have been kept together since weaning. Where individuals cannot be group-housed, consideration should be given to housing them in close visual contact.

#### 4.2. Enrichment

Suitable enrichment for rabbits includes roughage, hay blocks or chew sticks as well as an area for withdrawal. In floor pens for group housing, visual barriers and structures to provide refuges and look out behaviour should be provided. For breeding does nesting material and a nestbox should also be provided.

#### 4.3. Enclosures — dimensions and flooring

It is preferable for enclosures to be rectangular. A raised area should be provided within the enclosure. This raised area should allow the animal to lie and sit and easily move underneath, but should not cover more than 40 % of the floor space. While the enclosure height should be sufficient for the rabbit to sit upright without its ears touching the roof of the enclosure, this degree of clearance is not considered necessary for the raised area. If there are good scientific or veterinary reasons for not using a shelf then the enclosure should be 33 % larger for a single rabbit and 60 % larger for two rabbits. Wherever it is possible, rabbits should be kept in pens.

#### 4.3.1. Dimensions

Table B.1.

Rabbits over 10 weeks of age: Minimum enclosure dimensions and space allowances

Final body weight (kg)	Minimum floor area for one or two socially harmonious animals (cm²)	Minimum height (cm)
Less than 3	3 500	45
from 3 to 5	4 200	45
over 5	5 400	60

The table is to be used for both cages and pens. In cages a raised area should be provided (see Table B.4.). Pens should contain structures that subdivide the space to allow animals to initiate or avoid social contact. The additional floor area is  $3\,000\,\text{cm}^2$  per rabbit for the third, the fourth, the fifth and the sixth rabbit, while  $2\,500\,\text{cm}^2$  should be added for each additional rabbit above a number of six.

Table B.2.

Doe plus litter: Minimum enclosure dimensions and space allowances

Doe weight (kg)	Minimum enclosure size (cm²)	Addition for nestboxes (cm <sup>2</sup> )	Minimum height (cm)
Less than 3	3 500	1 000	45
from 3 to 5	4 200	1 200	45
over 5	5 400	1 400	60

At least three to four days before giving birth, does should be provided with an extra compartment or a nestbox in which they can build a nest. The nestbox should preferably be outside the enclosure. Straw or other nesting material should be provided. The enclosure should be designed so that the doe can move to another compartment or raised area away from her pups after they have left the nest. After weaning, the littermates should stay together in their breeding enclosure as long as possible. Up to eight littermates may be kept in the breeding enclosure from weaning until seven weeks old, and five littermates may be kept on the minimum floor area from eight to ten weeks of age.

Table B.3.

Rabbits less than 10 weeks of age: Minimum enclosure dimensions and space allowances

Age	Minimum enclosure size (cm²)	Minimum floor area per animal (cm²)	Minimum height (cm)
Weaning to 7 weeks	4 000	800	40
From 7 to 10 weeks	4 000	1 200	40

The table is to be used for both cages and pens. Pens should contain structures that subdivide the space to allow animals to initiate or avoid social contact. After weaning, the littermates should stay together in their breeding enclosure as long as possible.

Table B.4.

Rabbits: Optima dimensions for raised areas for enclosures having the dimensions indicated in Table B.1.

Age in Weeks	Final body weight (kg)	Optimum size (cm × cm)	Optimum height from the enclosure floor (cm)
Over 10	less than 3	55 × 25	25
	from 3 to 5	55 × 30	25
	over 5	60 × 35	30

To allow proper use of the raised area and of the enclosure as a whole the dimensions given above for the raised area size and height are optima, with very close minima and maxima (within  $10\,\%$  of optimum size). If there are scientific or veterinary justifications for not providing a raised area then the floor area should be  $33\,\%$  larger for a single rabbit and  $60\,\%$  larger for two rabbits, to facilitate the rabbit's locomotor activities and to enhance the opportunity to escape from a more dominant animal.

Where a raised area is provided for rabbits of less than 10 weeks of age, the optimum size of the raised area should be  $55 \times 25$  cm and the height above the floor should be such that the animals can make use of it.

## 4.3.2. Flooring

Wire floors should not be used without the provision of a resting area large enough to hold all the rabbits at any one time. Solid floors with bedding or perforated floors are preferable to grid or wire mesh floors.

## 4.4. Feeding

(See paragraph 4.6. of the General section)

## 4.5. Watering

(See paragraph 4.7. of the General section)

# 4.6. Substrate, litter, bedding and nesting material

(See paragraph 4.8. of the General section)

## 4.7. Cleaning

(See paragraph 4.9. of the General section)

## 4.8. Handling

(See paragraph 4.10. of the General section)

## 4.9. Humane killing

(See paragraph 4.11. of the General section)

#### 4.10. Records

(See paragraph 4.12. of the General section)

#### 4.11. Identification

(See paragraph 4.13. of the General section)

## C. SPECIES-SPECIFIC GUIDELINES FOR CATS

## 1. Introduction

The domestic cat is derived from the solitary African wild cat (*Felis silvestris libyca*), but has a strong tendency to learn social behaviour. With appropriate socialisation provided at an early age, such behaviour can be expressed both to conspecifics and man.

Good social interaction with humans encourage suitable temperament for subsequent studies. However, as cats lack dominance hierarchies and appear to lack mechanisms for reconciliation post-conflict, forming social relationships may be stressful. Visible signs that cats are stressed are not as straightforward to interpret as are those in dogs.

As cats are territorial and become attached to particular locations they are likely to be stressed by relocation. Cats are excellent climbers and utilise raised structures (e.g. shelves) extensively, both as vantage points and, when housed in groups, to maintain a distance from other cats.

#### 2. The environment and its control

## 2.1. Ventilation

(See paragraph 2.1. of the General section)

## 2.2. Temperature

Cats may be maintained within a wide temperature range provided that their welfare is not compromised. A temperature range of 15 °C to 21 °C should be maintained when precise control is required for cats under procedure (see paragraph 2.2.3 of the General Section).

As kittens have limited thermoregulatory control for around the first ten days of life, additional local heating should be provided during this period.

## 2.3. Humidity

It is considered unnecessary to control relative humidity, as cats can be exposed to wide fluctuations of ambient relative humidity without adverse effects.

## 2.4. Lighting

Holding of cats under the natural twenty-four-hour light-dark cycle is acceptable. Where the light part of the photoperiod is provided by artificial lighting, this should be within a range of ten to twelve hours daily.

If natural light is totally excluded, low level night lighting (5 to 10 lux) should be provided to allow cats to retain some vision and to take account of their startle reflex.

## 2.5. Noise

(See paragraph 2.5. of the General section)

#### 2.6. Alarm systems

(See paragraph 2.6. of the General section)

#### 3. Health

(See paragraphs 4.1. and 4.4. of the General section)

#### 4. Housing, enrichment and care

#### 4.1. Housing

Female cats and neutered cats of both sexes are generally sociable and are commonly held in groups of up to twelve. However, the establishment of groups of two or more such cats requires careful monitoring of the compatibility of all individuals in the group. Special care is needed when regrouping cats, introducing an unfamiliar cat to a group, housing un-neutered males in a group or maintaining cats in larger groups.

Where cats are normally group-housed, single-housing may be a significant stress factor. Therefore, cats should not be single-housed for more than twenty-four hours without justification on veterinary or welfare grounds. Single housing for more than twenty-four hours on experimental grounds should be determined in consultation with the animal technician and with the competent person charged with advisory duties in relation to the well-being of the animals.

Cats which are repeatedly aggressive towards other cats should be housed singly only if a compatible companion cannot be found. Social stress in all pair- or group-housed individuals should be monitored at least weekly using an established behavioural and/or physiological stress scoring system. This is especially important for un-neutered males.

Females with kittens under four weeks of age or in the last two weeks of pregnancy may be housed singly. During this time, consideration should also be given to allowing females which are normally group-housed to have access to their group e.g. by connecting kittening enclosures to the group housing animal enclosures.

The development of social behaviour in cats is profoundly affected by social experience between two and eight weeks of age. During this period it is particularly important that the cat has social contacts with other cats (e.g. litter mates) and with humans and is familiarised with environmental conditions likely to be encountered during subsequent use. Daily handling during this sensitive stage of development is a prerequisite for the social behaviour of the adult cat and it has been shown that a short period of handling even on the first day after birth is of importance as the young animals are already able to respond to scent and tactile stimulation.

All cats should have a period of play and general social interactions with humans on a daily basis, plus additional time for regular grooming. Particular attention should be paid to social enrichment for single-housed cats by providing additional human contact.

#### 4.2. Enrichment

Raised, part-enclosed structures should be provided (e.g. a bed with three walls and a roof on a shelf approximately one metre off the floor) to give the cats a view of their surroundings and, if pair- or group-housed, the opportunity to maintain a comfortable distance from other cats. There should be a sufficient number of these structures to minimise competition. Structures should be distributed within the enclosure so that animals can fully use the space available.

There should also be provision for the cats to seek refuge and privacy within their own enclosure and, in particular, away from the sight of cats in other enclosures. Vertical wooden surfaces should be provided to allow claw-sharpening and scent-marking.

Outside runs provide an environmental enrichment opportunity for cats in both breeding and user establishments and should be provided where possible.

Pseudo-predatory and play behaviour should be encouraged. A selection of toys should be available and these should be changed on a regular basis in order to ensure ongoing stimulation and avoid familiarity, which decreases the motivation to play.

## 4.3. Enclosures — dimensions and flooring

Enclosures, including the divisions between enclosures, should provide a robust and easy to clean environment for the cats. Their design and construction should seek to provide an open and light facility giving the cats comprehensive sight outside of their enclosure.

## 4.3.1. Dimensions

Table C.1.

Cats: Minimum enclosure dimensions and space allowances

	Floor (*) (m²)	Shelves (m²)	Height (m)
Minimum for one adult animal	1,5	0,5	2
For each additional animal add	0,75	0,25	_

<sup>(\*)</sup> Floor area excluding shelves.

The minimum space in which a queen and litter may be held is the space for a single cat, which should be gradually increased so that by four months of age litters have been re-housed to follow the above space requirements for adults. The normal age for weaning is seven to nine weeks.

Cats should never be forced to spend their entire lives outside and should always have access to an internal enclosure that meets all standards, including the minimum dimensions, detailed in these guidelines.

Areas for feeding and for litter trays should be not less than 0.5 metres apart and should not be interchanged.

Constraint in a space below the minimum requirement detailed above, such as in a metabolism cage or any similar type of housing for scientific purposes, may severely compromise the welfare of the animals. Such constraint should be for the minimum time and within a space that is as close as possible to that defined above and no less than that required for the animal to stretch fully horizontally and vertically, to lie down and turn around.

## 4.3.2. Flooring

The preferred flooring for cat enclosures is a solid continuous floor with a smooth non-slip finish. Additional enclosure furniture should provide all cats with a comfortable resting place.

Open flooring systems such as grids or mesh should not be used for cats. Where there is a justification for open flooring, great care should be taken in their design and construction in order to avoid pain, injury or disease and to allow the animals to manifest normal behaviours. Practical experience shows that metabolism cages are not always necessary as cat's urine and faeces can be collected directly from litter trays.

The quality and finish of the floor of an outside run need not be to the standard of the inside enclosure, providing it is easy to clean and not physically injurious to the cats.

# 4.4. Feeding

(See paragraph 4.6. of the General section)

#### 4.5. Watering

(See paragraph 4.7. of the General section)

## 4.6. Substrate, litter, bedding and nesting material

At least one litter tray of minimum dimension  $300 \times 400$  mm should be provided for every two cats and should contain a suitable absorbent and non-toxic litter or substrate material that is acceptable to and used by the cats. If urine and faeces are regularly deposited outside the trays, additional trays containing alternative substrates should be provided. If this is ineffective in pair- or group-housed cats, social incompatibility is indicated and cats should be removed from the group one at a time until the problem is resolved.

Sufficient beds should be provided for all cats and should be made of a suitable easy to clean material. These beds should contain bedding material such as polyester fleece or similar bedding material.

## 4.7. Cleaning

Each occupied enclosure should be cleaned at least daily; Litter trays should be emptied daily and litter material replaced.

Cleaning of enclosures should not result in cats becoming wet. When enclosures are hosed down, the cats should be removed from the enclosure to a dry place and returned only when it is reasonably dry.

#### 4.8. Handling

For cats, close contact with the persons caring for them is crucial, especially for single-housed cats.

#### 4.9. Humane killing

(See paragraph 4.11. of the General section)

#### 4.10. Records

(See paragraph 4.12. of the General section)

## 4.11. Identification

(See paragraph 4.13. of the General section)

## D. SPECIES-SPECIFIC GUIDELINES FOR DOGS

## 1. Introduction

The domestic dog (Canis familiaris) is an inquisitive and highly social animal which actively seeks information about its surroundings, reflecting the behaviour of its ancestors in the wolf family. Although much of the day is spent resting, the dog requires a complex physical and social environment during the active phase.

Bitches seek solitude in a quiet area for parturition and rearing of young.

As aggression is a significant risk, care is needed to maintain dogs in socially harmonious groups. The recommendations provided are for the beagle, the most commonly used breed. Account should be taken of individual breed characteristics if other breeds are used.

#### 2. The environment and its control

# 2.1. Ventilation

(See paragraph 2.1 of the General section)

## 2.2. Temperature

Dogs may be maintained within a wide temperature range provided that their welfare is not compromised. A temperature range of 15 °C to 21 °C should be maintained when precise control is required for dogs under procedure (see paragraph 2.2.3 of the General Section).

As puppies have limited thermoregulatory control in the first ten days or so of life, additional local heating should be provided within the whelping enclosure.

#### 2.3. Humidity

It is considered unnecessary to control relative humidity, as dogs can be exposed to wide fluctuations of ambient relative humidity without adverse effects.

## 2.4. Lighting

The holding of dogs under the natural twenty-four-hour light-dark cycle is acceptable. Where the light part of the photoperiod is provided by artificial lighting, this should be within a range of ten to twelve hours daily.

If natural light is totally excluded, low-level night lighting (5 to 10 lux) should be provided to allow dogs to retain some vision and to take account of their startle reflex.

#### 2.5. Noise

Noise in dog kennels can reach high levels which are known to cause damage to humans, and which could affect dogs' health or physiology. For these reasons it is important to consider methods of reducing noise in dog facilities. By addressing the dogs' behavioural needs in the facility design, the level of vocalisation may be decreased. Much of the noise is generated by the dogs' own vocalisations, but may also be generated by husbandry operations within the facility and ingress from outside sources. Any source of noise that may stimulate further dog barking should therefore be limited as far as possible. Penetration of external noise can be reduced by appropriate siting of the facility and by appropriate architectural design. Noise generated within the facility can be reduced by noise absorbent materials or structures. Expert advice on noise reduction should be taken when designing or modifying dog accommodation.

## 2.6. Alarm systems

(See paragraph 2.6. of the General section)

## 3. Health

(See paragraphs 4.1. and 4.4. of the General section)

## 4. Housing, enrichment and care

## 4.1. Housing

Dogs should be housed in socially harmonious groups within the animal enclosure, unless the scientific procedures or welfare requirements make this impossible. Special care is needed when regrouping dogs or introducing an unfamiliar dog to a group. In all cases, groups should be monitored for social compatibility on an ongoing basis.

Outside runs provide an environmental enrichment opportunity for dogs in both breeding and user establishments and should be provided where possible.

Single-housing of dogs for even short periods can be a significant stress factor. Therefore, dogs should not be single-housed for more than four hours without justification on welfare or veterinary grounds. Single-housing for more than four hours on experimental grounds should be determined in consultation with the animal technician and with the competent person charged with advisory duties in relation to the well-being of the animals.

In such circumstances, additional resources should be targeted to the welfare and care of these dogs. Additional human socialisation time, and visual, auditory and, where possible, tactile contact with other dogs, should be provided for all single-housed animals on a daily basis.

Unless contra-indicated on scientific grounds, single-housed dogs should be allowed to exercise in a separate area with, if possible, other dogs, and with staff supervision and interaction, on a daily basis.

Stud dogs should, wherever possible, be housed in socially harmonious pairs or groups or with bitches. Periparturient bitches should only be moved to the whelping enclosure between one and two weeks of expected parturition. While in the whelping enclosure they should have additional daily human contact.

Social behaviour in dogs develops between four and twenty weeks of age. During this period it is particularly important that the dog has social contacts with littermates, adult dogs (e.g. the bitch) and with humans, and is familiarised with conditions likely to be encountered during subsequent use. Daily handling during this sensitive stage of development is a prerequisite for the social behaviour of the adult dog and it has been shown that a short period of handling, even from the first day after birth onwards, is of importance as the young animals are already able to respond to scent and tactile stimulation.

#### 4.2 Enrichment

The design of indoor and outdoor enclosures should allow some privacy for the dogs and enable them to exercise some control over their social interactions.

Separate areas for different activities should be provided. This can be achieved by, for example, inclusion of raised platforms and pen sub-divisions.

Dog treats and toys afford welfare benefits to the animals, providing these are used sensibly and adequately monitored. As chewing is an important behaviour, items should be provided which meet this need.

The primary advantages of exercise are to allow additional opportunities for dogs to experience a complex and varied environment and to increase interaction with other dogs and humans. These will be particularly important where these needs cannot be fully met within the space provided by the animal enclosure. Therefore, unless contra-indicated on scientific or veterinary grounds, dogs should be removed to a separate area and allowed to exercise, with other dogs where possible, and with staff supervision and interaction, ideally on a daily basis.

## 4.3. Enclosures — dimensions and flooring

Animal enclosures, including the divisions between enclosures, should provide a robust and easy to clean environment for the dogs. In their design and construction they should seek to provide an open and light facility giving the dogs comprehensive sight of other dogs and staff, outside of their immediate animal enclosure.

#### 4.3.1. Dimensions

These guidelines are intended to encourage the social housing of dogs and to permit adequate environment enrichment. It should be noted that within this concept and strategy every encouragement is given to holding dogs in large and socially-harmonious groups both to increase the available floor space and to enhance socialisation opportunities.

Dogs should never be forced to spend their entire lives outside and should at all times have access to an internal enclosure that meets the standards for construction and environmental control detailed in these guidelines. The internal enclosure should represent not less than 50 % of the minimum space to be made available to the dogs, as detailed in Table D.1 below.

The space allowances detailed below are based on the requirements of beagles, but it should be noted that allowances significantly in excess may be required for giant breeds such as St Bernards or Irish wolfhounds. For breeds other than the laboratory beagle, space allowances should be decided in consultation with veterinary staff and the responsible authority.

Table D.1.

Dogs: Minimum enclosure dimensions and space allowances

Weight (kg)	Minimum enclosure size (m²)	Minimum floor area for one or two animals (m²)	For each additional animal add a minimum of (m²)	Minimum height (m)
Up to 20 over 20	4 4	4 8	2 4	2 2

Dogs that are pair or group housed may each be constrained to half the total space provided  $(2 \text{ m}^2 \text{ for a dog under } 20 \text{ kg}, 4 \text{ m}^2 \text{ for a dog over } 20 \text{ kg})$  while they are undergoing procedures as defined in Directive 86/609/EEC, if this separation is essential for scientific purposes. The period for which a dog should be so constrained should be minimised and should not in any event exceed four hours. This provision is made to encourage pair housing (particularly in toxicology studies) while at the same time allowing for the need to monitor feed intake and perform post-dosing observations.

Any further social or physical constraint, such as in a metabolism cage or physical restraint in a sling, may severely compromise the welfare of the animals. Constraint in a metabolism cage or any similar type of housing for scientific purposes should be within a space that is as close as possible to that defined above and no less than that required for the animal to stretch fully, lie down and turn around.

## 4.3.2. Nursing bitches and litters, and puppies up to 7,5 kg

A nursing bitch and litter should have the same space allowance as a single bitch of equivalent weight. The whelping pen should be designed so that the bitch can move to an additional compartment or raised area away from the puppies.

The normal weaning age for puppies is six to nine weeks.

Table D.2.

Dogs: Minimum enclosure dimensions and space allowances for post-weaned stock

Weight of dog (kg)	Minimum enclosure size (m²)	Minimum floor area/animal (m²)	Minimum height (m)
Up to 5	4	0,5	2
over 5 to 10	4	1,0	2
over 10 to 15	4	1,5	2
over 15 to 20	4	2	2
over 20	8	4	2

## 4.3.3. Flooring

The preferred flooring for dog accommodation is a solid continuous floor with a smooth non-slip finish. All dogs should be provided with a comfortable, solid resting area, for example, by the use of enclosure furniture such as raised beds or platforms.

Open flooring systems such as grids or mesh should not be used for dogs. Where there is a justification for open flooring, great care should be taken in their design and construction in order to avoid pain, injury or disease and to allow the animals to manifest normal behaviours. If any welfare problems do arise which are related to the flooring, veterinary advice should be sought and, if necessary, the dogs relocated onto solid flooring.

Pre-weaned puppies and peri-parturient and suckling bitches should not be kept in an open floor system.

The quality and finish of the floor of an outside run need not be to the standard of the inside enclosure, providing it is easily cleanable and not injurious to the dogs.

## 4.4. Feeding

(See paragraph 4.6. of the General section)

## 4.5. Watering

(See paragraph 4.7. of the General section)

## 4.6. Substrate, litter, bedding and nesting material

When dogs are held on solid floors, some litter or substrate material facilitates cleaning and minimises the necessity for regular washing or hosing down.

Peri-parturient and suckling bitches should be provided with a bed and bedding material to support whelping and the nursing of puppies. Puppies also benefit from the provision of bedding materials, as may certain breeds such as the greyhound.

## 4.7. Cleaning

Each occupied enclosure should be cleaned at least daily. All excreta and soiled materials should be removed from all areas used by dogs at least daily, and more frequently if necessary.

Wet cleaning by hosing down of enclosures should be carried out as necessary but should not result in the dogs becoming wet. When enclosures are hosed down, the dogs should be removed from the enclosure to a dry place and returned only when it is reasonably dry.

## 4.8. Handling

(See paragraph 4.1. above and paragraph 4.10. of the General section)

#### 4.9. Humane killing

(See paragraph 4.11. of the General section)

## 4.10. Records

(See paragraph 4.12. of the General section)

## 4.11. Identification

(See paragraph 4.13. of the General section)

## E. SPECIES-SPECIFIC GUIDELINES FOR FERRETS

## 1. Introduction

Ferrets (Mustela putorius furo) are carnivores which under natural conditions feed on small mammals, birds, fish and invertebrates. They have complex hunting behaviour and tend to hoard food, but will not eat decayed matter.

Although in the wild the ferret is generally a solitary animal, there seem to be welfare benefits if they are housed in socially harmonious groups in captivity. Ferrets normally live in burrows, and thus in captivity appreciate the provision of materials, such as tubes in which they can crawl and play games.

Ferrets usually breed once a year, mating in the spring. Male animals are hostile to, and will fight vigorously with, unfamiliar males during the breeding season. As a consequence, at this time single housing of males may prove necessary.

The ferret is an intelligent, inquisitive, playful and agile animal, and this should be taken into account in the design of the accommodation and when handling. A complex, escape-proof enclosure is required which provides opportunities to the ferret to exhibit a wide behavioural repertoire.

## 2. The environment and its control

#### 2.1. Ventilation

(See paragraph 2.1. of the General section)

## 2.2. Temperature

Ferrets should be maintained in the temperature range of 15 °C to 24 °C.

As ferrets do not have well-developed sweat glands, to avoid heat exhaustion they should not be exposed to high temperatures.

#### 2.3. Humidity

It is considered unnecessary to control or record relative humidity as ferrets can be exposed to wide fluctuations of ambient relative humidity without adverse effects.

#### 2.4. Lighting

The light source and type should not be aversive to the animals and particular care should be taken with ferrets, especially if albino, housed in the top tier of tiered racking systems.

Holding of ferrets under the natural twenty-four-hour light-dark cycle is acceptable.

Where the light part of the photoperiod is provided by artificial lighting, this should be for a minimum of eight hours daily and should generally not exceed sixteen hours daily.

However, it should be noted that for manipulation of the reproductive cycle variation in the light-dark cycles is necessary (e.g. the light part of the photoperiod can vary from six to sixteen hours).

If natural light is totally excluded, low level night lighting should be provided to allow animals to retain some vision and to take account of their startle reflex.

#### 2.5. Noise

Lack of sound or auditory stimulation can be detrimental and make ferrets nervous. However, loud unfamiliar noise and vibration have been reported to cause stress-related disorders in ferrets and should be avoided. It is important to consider methods of reducing sudden or unfamiliar noise in ferret facilities, including that generated by husbandry operations within the facility and also by ingress from outside sources. Ingress of noise can be controlled by appropriate siting of the facility and by appropriate architectural design. Noise generated within the facility can be controlled by noise absorbent materials or structures. Expert advice should be taken when designing or modifying accommodation.

#### 2.6. Alarm systems

(See paragraph 2.6. of the General section)

## Health

(See paragraphs 4.1. and 4.4. of the General section)

# 4. Housing, enrichment and care

#### 4.1. Housing

Animals should be kept in socially harmonious groups unless there are scientific or welfare justifications for single housing.

During the breeding season, adult males may need to be maintained singly to avoid fighting and injury. However, males can be maintained successfully in groups at other times.

Pregnant females should be housed singly only during late pregnancy, no more than two weeks prior to parturition.

Separation of animals that are normally group-housed can be a significant stress factor. Where this is for a period of more than twenty-four hours, it should be regarded as severely compromising the welfare of the animals. Therefore, ferrets should not be single-housed for more than twenty-four hours without justification on veterinary or welfare grounds. Single housing for more than twenty-four hours on experimental grounds should be determined in consultation with the animal technician and with the competent person charged with advisory duties in relation to the well-being of the animals.

Where animals are single-housed, whether for scientific or welfare reasons, additional resources should be targeted to the welfare and care of these animals. Additional human socialisation time, and visual, auditory and, where possible, tactile contact with other ferrets should be provided for all single-housed animals on a daily basis.

The social behaviour of ferret should be taken into account by providing regular interaction with other ferrets through group housing and regular handling. In general, ferrets seem to benefit from such regular and confident handling and this should be encouraged, as it results in better quality and more sociable animals.

Social behaviour in ferrets develops at an early age and it is important that the young ferret has social contacts with other ferrets (e.g. litter-mates) and with humans (e.g. animal caretakers). Daily handling during this sensitive stage of development is a prerequisite for the social behaviour of the adult ferret. It is reported that the more frequent the interaction, the more placid the animal will become, and this interaction should be continued through into adult life.

#### 4.2. Enrichment

The design of the ferret enclosure should meet the animals' species- and breed-specific needs. It should be adaptable so that innovation based on new understanding may be incorporated.

The design of the enclosure should allow some privacy for the ferrets and enable them to exercise some control over their social interactions.

Separate areas for different activities, such as by raised platforms and pen sub-divisions, should be provided in addition to the minimum floor space detailed below. Where nesting boxes are provided, these should be designed to contain the young ferrets within the nest.

Provision of containers and tubes of cardboard or rigid plastic, and paper bags, stimulates both investigative and play behaviour. Water baths and bowls are used extensively by ferrets.

## 4.3. Enclosures — dimensions and flooring

These guidelines are intended to encourage the social housing of ferrets and to permit adequate enrichment of the environment. It should be noted that within this concept and strategy every encouragement is given to holding ferrets in large and socially harmonious groups both to increase the available floor space and to enhance the socialisation opportunities.

Animal enclosures, including the divisions between enclosures, should provide an easy to clean and robust environment for the ferrets. Their design and construction should seek to provide an open and light facility giving the ferrets comprehensive sight of other ferrets and staff, outside of their immediate animal enclosure. There should also be provision for the ferrets to seek refuge and privacy within their own enclosure and, in particular, away from the sight of ferrets in other enclosures.

As ferrets have a remarkable ability to escape, the design of the enclosure should be such that the animal is unable to escape or to injure itself should any such attempt be made.

The recommended minimum height of the enclosure should be 50 cm. The ferret enjoys climbing and this height facilitates provision of suitable enrichment. The floor space should provide an adequate area for movement and to allow the animal the opportunity to select sleeping, eating and urination/defecation areas. In order to provide enough space for environmental complexity, no animal enclosure should be less than  $4\,500$  cm $^2$ . Minimum space requirements for each ferret are as follows:

Table E.1.

Ferrets: Minimum enclosure dimensions and space allowances

	Minimum enclosure size (cm²)	Minimum floor area per animal (cm²)	Minimum height (cm)
Animals up to 600 g	4 500	1 500	50
Animals over 600 g	4 500	3 000	50
Adult males	6 000	6 000	50
Jill and litter	5 400	5 400	50

Animal enclosures should be of a rectangular shape rather than square, to facilitate locomotor activities.

Constraint in less than the above space requirements for scientific purposes, such as in a metabolism cage, may severely compromise the welfare of the animals

#### 4.3.1. Flooring

ΕN

The flooring for ferret accommodation should be a solid continuous floor with a smooth non-slip finish. Additional enclosure furniture such as beds or platforms should provide all ferrets with a warm and comfortable resting place.

Open flooring systems such as grids or mesh should not be used for ferrets.

## 4.4. Feeding

(See paragraph 4.6. of the General section)

## 4.5. Watering

(See paragraph 4.7. of the General section)

## 4.6. Substrate, litter, bedding and nesting material

Bedding material is required for all ferrets. In addition, nesting materials such as hay, straw or paper should be provided. Deep litter systems are considered to provide additional enrichment.

It is good practice to use some litter or substrate material at least to facilitate cleaning and minimise the necessity to wash/hose down regularly.

## 4.7. Cleaning

Wet cleaning by hosing down of animal enclosures should not result in ferrets becoming wet. When animal enclosures are hosed down, the ferrets should be removed from the enclosure to a dry place and returned only when it is reasonably dry.

Ferrets tend to defecate against a vertical surface in one area of the enclosure. Provision of a litter tray may be beneficial and reduce the frequency of cleaning required for the remainder of the enclosure.

All excreta and soiled materials should be emptied at least daily, and more frequently if necessary, from litter trays and/or removed from all other areas used by the animals as a toilet.

Frequency of cleaning of the remainder of the enclosure should be determined on factors such as stocking density, enclosure design and stage of breeding e.g. periparturient period.

## 4.8. Handling

(See paragraph 4.10. of the General section)

## 4.9. Humane killing

(See paragraph 4.11. of the General section)

#### 4.10. Records

(See paragraph 4.12. of the General section)

# 4.11. Identification

(See paragraph 4.13. of the General section)

### F. SPECIES-SPECIFIC GUIDELINES FOR NON-HUMAN PRIMATES

### a. General considerations

### 1. Introduction

Keeping non-human primates in the laboratory creates a number of problems which are not shared with other commonly used laboratory mammals. Non-human primates are not domesticated, but are wild animals; most are also arboreal. Their wild status means that they are more alert than domesticated species and thus are highly reactive to any unfamiliar and alarming stimuli. Unlike domesticated species, they have not been selected for friendliness to humans and low aggression. Early friendly contact between infants and care-givers will result in a less fearful animal, as the animals learn that familiar humans do not constitute a threat, but the animals will retain most of the attributes of their wild conspecifics. In contrast to non-arboreal laboratory mammals, the flight reaction of non-human primates from terrestrial predators is vertical, rather than horizontal; even the least arboreal species seek refuge in trees or on cliff faces. As a result, enclosure height should be adequate to allow the animal to perch at a sufficiently high level for it to feel secure. The structural division of space in primate enclosures is of paramount importance. It is essential that the animals should be able to utilise as much of the volume as possible because, being arboreal, they occupy a three-dimensional space. To make this possible, perches and climbing structures should be provided.

In addition to their wild nature and climbing habits, non-human primates have advanced cognitive capabilities and complex foraging and social behaviour. As a result, they require complex, enriched environments to allow them to carry out a normal behavioural repertoire. The group structure, however, should be such that normal behaviours indicative of distress or pain or those likely to result in injury are kept to a minimum.

Non-human primates used for scientific research should be captive-bred and, where practicable, reared on site to avoid transport stress. Captive-bred animals are of known age, parentage and health status and have been reared under standardised husbandry practices. Where non-human primates are to be imported they should, whenever possible, be obtained as offspring from established breeding colonies with high welfare and care standards. They should be free from zoonotic diseases. Wild caught animals should only be used in exceptional circumstances as they present health hazards to staff, have unknown histories and are likely to be more afraid of humans. In some instances there can be a significant mortality among the animals at the trapping site and during transfer to the source country holding site.

Additional details are provided for the commonly bred and used laboratory species. Further advice on requirements for other species (or if behavioural or breeding problems occur) should be sought from experienced primatologists and care staff to ensure that any particular species needs are adequately addressed.

# 2. The environment and its control

# 2.1. Ventilation

(See paragraph 2.1. of the General section)

# 2.2. Temperature

As in captivity the animals have restricted opportunities for natural behavioural means of coping with climatic change, the ranges specified for laboratory animals will not necessarily reflect those which they experience in nature. Generally the ranges will be those which are optimal for the animals and comfortable for staff. Where outdoor enclosures are in use, it is essential to provide shelter from inclement weather for all individuals and continuous access to adequate heated indoor accommodation. This is of particular importance in breeding colonies with extensive outdoor enclosures to reduce the risk of frostbite and loss of neonates in the winter months.

# 2.3. Humidity

Although some non-human primates live in tropical rain forests, where humidity is high, and others in arid regions, it is not necessary for this to be replicated in the laboratory for established colonies. In general, humidity levels of 40 to 70 % relative humidity are comfortable for both animals and care staff. Care should be taken (see individual species) not to expose the animals to humidity which is too low and prolonged exposure outside this range should be avoided, particularly for New World monkeys, which may be susceptible to respiratory problems.

# 2.4. Lighting

Most laboratory non-human primates should have a 12 hour/12 hour light/dark cycle. Simulated dawn and dusk lighting may be beneficial for some species. For the nocturnal species, such as *Aotus trivirgatus*, the cycle should be modified so that dim red light is used during part of the normal working day to allow the animals to be observed during their active periods, and also to enable routine husbandry tasks to be carried out safely. Whenever possible, rooms housing non-human primates should be provided with windows, since they are a source of natural light and can provide environmental enrichment.

### 2.5. Noise

Restful background sound such as music or radio programmes provided during the day can act as a form of environmental enrichment and help to screen out sudden loud noises but it should not be provided permanently. Music may also have a calming effect on the animals in times of stress. For most species, satisfactory sound levels will be the same as those recommended for staff, but some species such as callitrichids can also hear ultrasound, so this should be taken into account. The level of background noise should be kept low and should only exceed 65 dBA for short periods.

# 2.6. Alarm systems

Most higher non-human primates have similar hearing to humans; to avoid frightening the animals sirens should be avoided. An appropriate alternative would be to use flashing lights visible to staff in all rooms.

### 3. Health

Though the use of captive-bred animals should ensure that they are in good health and do not pose a risk of infection to staff or other non-human primates in the premises, all newly acquired animals should arrive with full health certification and be quarantined on arrival. During this period their health should be closely monitored and further serological, bacteriological and parasitological tests should be performed by competent laboratories as required.

All non-human primates in the colony should be under expert veterinary control and submitted to periodical diagnostic tests. Their close affinity to humans results in susceptibility to a number of diseases and parasites that are common to both and occasionally life threatening to the other. It is, therefore, of vital importance that there is also regular medical screening of the staff. Any member of staff posing a potential health risk to the animals should not have contact with the animals. Particular care should be taken when dealing with animals which may be contaminated by pathogens transmissible to humans. Staff should be informed, and measures taken to minimise the risk of infection. Lifetime health records should be kept for each animal. The investigation of unexpected morbidity and mortality should be thorough, having regard for potential zoonotic diseases, and be entrusted to competent personnel and laboratories.

Non-human primates from different geographical areas should be strictly separated from each other until their health status has been clarified.

In outdoor enclosures vermin control is of particular importance.

# 4. Housing, enrichment and care

# 4.1. Housing

A person competent in the behaviour of non-human primates should be available for advice on social behaviour, environmental enrichment strategies and management.

Because the common laboratory non-human primates are social animals, they should be housed with one or more compatible conspecifics. To ensure harmonious relations, it is essential that the group composition of laboratory non-human primates should be appropriate. Compatibility, and hence group composition, in terms of the age and sex of its members depends on the species. In creating groups, the natural social organisation of the species should be taken into account. In confined conditions, however, where the space for extended chases or the emigration of social rejects is not available, the natural age and sex composition of troops may be inappropriate, modifications to group structure may be required. For example, a harem structure may be substituted for the natural multi-male, multi-female troop in macaques. Experimental protocol may also determine group composition, for example, single-sex or same age groups. Visual barriers, which allow the animals to be out of sight of one another, are important in group housing and multiple escape routes provide opportunities to avoid attacks and also prevent dominant individuals from restricting access of subordinates to other parts of the enclosure.

Careful monitoring of animals is necessary following grouping or mixing, and a programme of action should be in place for managing and minimising aggressive interaction.

Where animals are housed in same-sex groups, it is best to avoid housing the two sexes in close proximity, as this can sometimes lead to the males becoming aggressive. The only exceptions to social housing should be either for veterinary reasons or where an experimental protocol demands it to ensure good science. Single housing should only be allowed for as short a time as possible, under close supervision, where there is a justification on veterinary or welfare grounds. Single housing on experimental grounds should be determined in consultation with the animal technician and with the competent person charged with advisory duties in relation to the well-being of the animals. In such circumstances, additional resources should be targeted to the welfare and care of these animals. For experimental animals, where housing them in large groups is not possible, keeping them in same-sex compatible pairs is probably the best social arrangement.

Where socially housed animals need to be separated for a period of time, for example, for dosing, care and vigilance should be exercised on re-introduction as the social organisation in the group may have changed and the animal may be attacked. Possible solutions include confinement of this animal to an individual enclosure attached to, or within, the main living area or separation of all individuals briefly followed by re-introduction of the whole group simultaneously.

# 4.1.1. Breeding

The sex ratio and numbers of animals in a breeding colony will depend on the species involved. It is important to ensure that both space and complexity are adequate to prevent the intimidation of individuals, particularly low-ranking females and young. In polygamous species, the sex ratio should ensure that the majority of females are mated and give birth to live offspring. Where there is more than one male in the group, care should be taken to ensure that the males are compatible. Monogamous species will be bred in family groups with a breeding pair and two or more sets of their offspring.

For future breeding animals, it is important that the young grow up in stable social groups, preferably their natal group, with their mothers. This ensures that their parenting skills and social interactions within a hierarchical structure develop adequately.

Animals will normally successfully rear single or twin offspring without intervention. However, a management policy for rejected infants is required to minimise suffering in these animals.

# 4.1.2. Separation from the mother

Young animals have a slow postnatal development lasting several years in cercopithecoids with a period of dependency on their mothers lasting until they are 8 to 12 months old, depending on the species. During this period they learn about their environment under the mother's protective vigilance and socialise through interactions with a diversity of social partners.

They also learn parenting skills by interacting with infants or even helping to care for them. Separation of infants from a colony causes distress to the mother and infant at the time. It is therefore preferable to leave them in their natal colony until they have become independent. Should they, for their own welfare, have to be weaned or separated earlier, it is advisable to incorporate them into a well organised group to avoid damage to their social development, behaviour, physiology and immune competence. The appropriate age ranges for weaning will depend on the species.

### 4.2. Enrichment

The environment should enable the animal to carry out a complex daily programme of activity. The precise features of the living quarters, however, will vary according to species, due to differences in natural behaviour. The enclosure should allow the animal to adopt as wide a behavioural repertoire as possible, provide it with a sense of security, and a suitably complex environment to allow the animal to run, walk, climb and jump. Materials providing tactile stimuli are also valuable. Opportunities for the animals to have some control over the environment should be provided. Some novelty should also be introduced at intervals, which can include for example minor changes in the conformation or arrangement of enclosure furniture and feeding practices.

# 4.3. Enclosures — dimensions and flooring

Non-human primates should be housed in such a way that they do not exhibit abnormal behaviour and are able to display a satisfactory range of normal activities.

The following factors will determine the enclosure dimensions for a given species:

- the adult size of the animal (juvenile animals, though smaller, are usually more active than adults, thus
  requiring similar space allowances for physical development and play), and
- sufficient space to provide a complex and challenging environment, and
- the size of group to be accommodated.

### 4.3.1. Dimensions

The following principles should apply to the housing of all species of non-human primates:

- enclosures should be of adequate height to allow the animal to flee vertically and sit on a perch or a shelf, without its tail contacting the floor,
- the animal should be able to display a normal locomotor and behavioural repertoire,
- there should be room for suitable environmental enrichment,
- apart from exceptional circumstances, the animal should not be singly housed,
- enclosures should not be arranged in two or more tiers vertically.

### 4.3.2. Outdoor enclosures

Where possible, non-human primates should have access to outdoor enclosures. These are commonly used for breeding larger non-human primates. They have the advantage for the animals that they can include many features of the natural environment and are also useful for holding stock or experimental animals where close climatic control is not required and outdoor temperatures are suitable. Outdoor enclosures are usually constructed of metal, but other materials, including wood, can be used providing it is suitably weather-proofed. Some types of wood are approved by toxicologists provided that a certificate of analysis is available. Wood is easily maintained or replaced, can be custom-built on site and provides a quieter and more natural material. To protect the structural integrity of a wooden enclosure, the framework should either be of a type of wood which the animals will not chew or protected with mesh and a non-toxic treatment. The base of the enclosure can be of concrete or natural vegetation. Concrete-floored enclosures can be covered with a suitable non-toxic substrate. Either part of the outdoor enclosure should be roofed, to allow the animals to be outside in wet weather and to provide protection from the sun or, alternatively, shelters can be provided. Where outdoor enclosures are provided, the non-human primates will utilise them, even in the winter. However, heated indoor enclosures should be provided. It is recommended that the minimum size for an indoor enclosure should meet the minimum values specified to ensure that the animals are not overcrowded in inclement weather. As outdoor enclosures represent additional space, there is no need to set minimum dimensions for these. Where different enclosures are connected, for example outdoor and indoor, more than one connecting door should be provided to prevent subordinates being trapped by more dominant animals.

# 4.3.3. Indoor housing

Although indoor enclosures will commonly be constructed of metal, other materials, such as wood, laminates and glass have been used successfully and provide a quieter environment.

As height is a critical feature of the enclosure, all non-human primates should be able to climb, jump and occupy a high perch. The walls can include mesh to allow climbing but sufficient diagonal branches or perches should also be provided to allow all animals to sit on them simultaneously. Where mesh is used, care should be taken to ensure that it is of a type which could not lead to injury through animals having their limbs trapped.

Solid floors have the advantage that they can be covered with a substrate in which food can be scattered to encourage foraging. Non-human primates require space for activity, but may need to be confined in smaller home enclosures for short periods of time when justified on veterinary or experimental grounds. Smaller volumes can be created by partitioning the main enclosure using dividers and/or a mobile back to the enclosure, having a cage within the home enclosure, two linked units, or attaching experimental enclosures to a larger exercise enclosure. These methods of confining experimental animals all have the advantage that animals have access to a satisfactory living environment and social companions, allowing however separation for feeding, cleaning and experimental purposes, such as dosing and blood sampling.

Should single-housing in a small enclosure be necessary, because of a special experimental paradigm, the duration and extent of confinement should be justified by the experimenter, balancing the likely effect on the well-being of the animal against the scientific value and requirements of the experiment. Such restrictions should be reviewed by scientists, animal technicians and those competent persons charged with advisory duties in relation to the well-being of the animals.

More space for activity can be provided by keeping non-human primates in large groups, rather than pairs. Individuals can be isolated by training (see paragraph 4.8 below) or running the group through a race with a trap in it.

The additional guidelines provide minimum recommended enclosure sizes for the different species.

### 4.4. Feeding

Presentation and content of the diet should be varied to provide interest and environmental enrichment. Scattered food will encourage foraging, or where this is difficult food should be provided which requires manipulation, such as whole fruits or vegetables, or puzzle-feeders can be provided. Foraging devices and structures should be designed and situated to minimise contamination. Vitamin C is an essential component of the primate diet. New World monkeys require adequate quantities of vitamin D<sub>3</sub>. As the enrichment feeding may lead to preferences, to ensure that the animals receive a balanced diet it is advisable to feed the standard diet first thing in the morning when the animals are hungry and have no alternative. The food can be scattered to ensure that it is not monopolised by dominant individuals. A varied diet should not be provided if it is likely to have disturbing effects on experimental results. However, in such circumstances variation can be introduced in the form of nutritionally standard diets available in different shapes, colours and flavours.

# 4.5. Watering

(See paragraph 4.7. of the General section)

# 4.6. Substrate, litter, bedding and nesting material

Some non-human primates, for example some prosimians, require nesting material, for example wood wool, dry leaves or straw. Non-toxic substrates such as wood chips, wood granulate with a low dust level or shredded paper are valuable to promote foraging in indoor enclosures. Grass, herbage wood chip or bark chip are suitable for outdoor facilities.

# 4.7. Cleaning

(See paragraph 4.9. of the General section)

# 4.8. Handling

Various methods of restraint are employed in handling non-human primates, ranging from enclosures with sliding partitions, through netting, holding the animals manually, to using a dart to tranquillise them. Although non-human primates dislike being handled and are stressed by it, training animals to cooperate should be encouraged, as this will reduce the stress otherwise caused by handling. Training the animals is a most important aspect of husbandry, particularly in long-term studies. It has a dual advantage in providing the animal with an intellectual challenge and making work more rewarding for the care-giver. Non-human primates will respond to aural and visual stimuli, and by using simple reward systems, training can often be employed to encourage the animals to accept minor interventions, such as blood sampling.

The response of individuals to training and procedures should be regularly reviewed, as some animals may be particularly difficult or non-responsive and in such cases, careful consideration should be given to their continued use.

Though animals can be trained to accomplish tasks, attention should be paid to appropriate recovery periods when subjected to repeated experiments.

### 4.9. Humane killing

(See paragraph 4.11. of the General section)

# 4.10. Records

Individual records containing detailed information for each animal should be maintained. These should include: species, sex, age, weight, origin, clinical and diagnostic information, present and previous housing system, history of experimental use and any other information relevant for management and experimental procedures, such as reports on their behaviour or status, and favoured social companions/social relationship.

# 4.11. Identification

All non-human primates in a facility should be identified with a permanent and unique laboratory identification code before weaning. Individual animals can be identified visually by using properly fitted necklaces with attached medallions or tattoos for large species. Microchips can be injected into accessible sites (the wrist for larger animals or scruff of the neck for smaller species. As it is important to be able easily to distinguish animals, some laboratories successfully use names for the animals, as these can easily be used to identify dominant and subordinate animals, and are considered by some to encourage the care staff to increase their respect for the non-human primates.

# 5. Training of personnel

Staff should be trained in the management, husbandry and training of animals under their care. For animal carers and scientists working with non-human primates, training should include species-specific information. This should include the biological and behavioural characteristics and requirements of the species, environmental enrichment, methods used for the introduction and removal of animals and social dynamics. Training should also include information on the health and safety of staff working with non-human primates including zoonotic disease risk, and management.

# 6. **Transport**

Animals should, where possible, be transported in compatible pairs. However, adult animals may need to be transported singly.

# b. Additional guidelines for housing and care of marmosets and tamarins

### 1. **Introduction**

Marmosets (*Callithrix spp.*) are small, highly arboreal, South American diurnal non-human primates. In the wild they have home ranges of 1 to 4 hectares where they live in extended family groups of three to fifteen animals consisting of a breeding pair and their offspring. Females produce litters twice a year (normally twins and in captivity, not infrequently, triplets) and all group members take care of the offspring. Reproductive inhibition of the subordinate females by the dominant occurs due to hormonal and behavioural mechanisms. Marmosets are frugivore-insectivore and are specialised in gum-tree gouging and gum feeding; however, in captivity they would gouge and scent-mark other hardwoods. Foraging and feeding occupy up to 50 % of the time available. Marmosets and tamarins can live for up to 15 to 20 years in captivity.

Tamarins (*Saguinus spp.*) are similar to marmosets in many respects. They are found in South and Central America, but are slightly larger animals and have larger home ranges, varying from 30 to 100 hectares. The larger home ranges of tamarins are related to more frugivorous diets, while they do not gouge, and eat gum only when readily accessible.

Most marmosets and tamarins show reluctance to descend to the ground and frequently scent-mark their environment.

### 2. The environment and its control

### 2.1. Ventilation

(See paragraph 2.1. of the General section)

# 2.2. Temperature

Marmosets and tamarins should be maintained in a temperature range of 23 °C to 28 °C, although levels slightly higher are acceptable due to the tropical nature of the animals.

### 2.3. Humidity

Humidity levels of 40 to 70 % should be provided, although the animals will tolerate relative humidity levels higher than 70 %.

### 2.4. Lighting

A photoperiod of no less than twelve hours of light is recommended. The lighting source should illuminate uniformly the holding room. However, within the animal enclosures, a shaded area should always be provided.

### 2.5. Noise

Special consideration should be given to minimise exposure to ultra-sound, which is within the hearing range of marmosets and tamarins.

### 2.6. Alarm system

(See paragraph 2.6. of the General considerations for non-human primates)

# 3. Health

(See paragraph 3 of the General considerations for non-human primates)

### 4. Housing, enrichment and care

# 4.1. Housing

Marmosets and tamarins should be housed in family groups consisting of unrelated male-female pairs and one or more sets of offspring. Groups of stock animals should consist of compatible same-sex peer individuals or juveniles. Care should be taken when grouping unrelated adult individuals of the same sex since overt aggression may occur.

During experiments, marmosets and tamarins can generally be kept with a compatible same-sex animal (twins, parent/offspring) or in male-female pairs, using contraception. When experimental procedures or veterinary care require single housing, the duration should be minimised and the animals should remain in visual, auditory and olfactory contact with conspecifics.

Breeding pairs should be formed only when the animals are aged about 2 years. In family groups, the presence of the mother will inhibit the ovulatory cycle in her female offspring. New pairs intended for breeding should not be kept close to the parental family since reproduction may be inhibited.

The appropriate age of weaning will depend on the intended use of the animals but should not be earlier than 8 months of age. When animals are to be used as breeders, they should remain in the family group until at least 13 months of age in order to acquire adequate rearing experience.

### 4.2. Enrichment

The natural behaviour of marmosets and tamarins indicates that the captive environment should provide some degree of complexity and stimulation, factors which are more valuable than simply increasing enclosure dimensions to promote species-typical behaviour. Furniture of natural or artificial materials (for example, wood, PVC) should include: perches, platforms, swings, ropes. It is important to provide a certain degree of variability in orientation, diameter and firmness to allow the animals to perform appropriate locomotor and jumping behaviours. Wooden perches allow marmosets and tamarins to express their natural behaviour of gnawing

followed by scent-marking. In addition, a comfortable secure resting area such as nest boxes should be included since they are used for resting, sleeping and hiding in alarming situations. Though visual contact between family groups is normally stimulating for the animals, opaque screens and/or increasing the distance between enclosures in order to avoid territorial interaction may be needed in some cases, and in particular for certain callitrichid species. Foraging devices, which stimulate the natural behaviour of the animals, should be suspended or presented in the upper part of the enclosure, in consideration of the reluctance of the animals to descend to ground level. Wood chips as a substrate will encourage foraging of spilled food at the floor area. In general, the inclusion in the lower part of the enclosure of structural elements and enrichment devices will promote a wider and more diversified use of the space. For marmosets, which are specialised in tree-gnawing to obtain gum, sections of dowel drilled with holes and filled with gum arabic have proved very beneficial.

# 4.3. Enclosures — dimensions and flooring

For marmosets and tamarins the volume of available space and the vertical height of the enclosure are more important than floor area, due to the arboreal nature and the vertical flight reaction of these species. The minimum dimensions and design of the enclosure should take into account the purpose for which the animals are maintained (breeding, stock, short or long experiments) and enable the inclusion of sufficient devices for improving the environmental complexity.

Table F.1.

Marmosets and Tamarins: Minimum enclosure dimensions and space allowances.

	Minimum floor area of enclosures for 1 (*) or 2 animals plus offspring up to 5 months old (m²)	Minimum volume per additional animal over 5 months (m³)	Minimum enclosure height (m) (**)
Marmosets	0,5	0,2	1,5
Tamarins	1,5	0,2	1,5

<sup>(\*)</sup> Animals should only be kept singly under exceptional circumstances (see paragraph 4.1). (\*\*) The top of the enclosure should be at least 1,8m from the floor.

# 4.4. Feeding

Marmosets and tamarins require a high protein intake and since they are unable to synthesise vitamin  $D_3$  without access to UV-B radiation, the diet must be supplemented with adequate levels of vitamin  $D_3$ .

# 4.5. Watering

(See paragraph 4.7. of the General section)

# 4.6. Substrate, litter, bedding and nesting material

(See paragraph 4.6. of the General considerations for non-human primates)

# 4.7. Cleaning

Marmosets and tamarins frequently scent-mark their environment and the total removal of familiar scents may cause behavioural problems. Alternate cleaning and sanitation of the enclosure and the enrichment devices retains some of the territorial scent-marking and has beneficial effects on the psychological well-being of the animals, reducing over-stimulated scent-marking.

# 4.8. Handling

Regular handling and human contact are beneficial for improving the animals' habituation to monitoring and experimental conditions and facilitate training to cooperate with some procedures. When capture and transport of the animals are required, nest boxes can be used to reduce handling stress.

# 4.9. Humane killing

(See paragraph 4.11. of the General section)

### 4.10. Records

(See paragraph 4.10. of the General considerations for non-human primates)

### 4.11. Identification

(See paragraph 4.11. of the General considerations for non-human primates)

# 5. Training of personnel

(See paragraph 5 of the General considerations for non-human primates)

# 6. Transport

(See paragraph 6 of the General considerations for non-human primates)

# c. Additional guidelines for housing and care of squirrel monkeys

### 1. **Introduction**

Squirrel monkeys (Saimiri spp.) inhabit the tropical rain forests of the South American continent at various altitudes. There are various regional subspecies, the two most important are known as S. sc. boliviensis (black headed) and S. sc. sciureus (olive). In addition to differences in coat colour and face masks they also have some minor variations in behavioural characteristics. Body weight of adults ranges from 600 to 1 100 g, with males being distinctly heavier than females. Standing upright, adult animals reach about 40 cm body length. They are typically arboreal animals living at different levels of the canopy, depending on environmental temperature. They do, however, descend to the ground to look for food and, and in the case of young animals, to play. When in danger, they flee to a high level. When travelling they may take leaps depending on the density of the canopy. In the wild they live in fairly large groups in which females and young animals live together with a dominant breeding male, whereas adult males that are not in breeding condition remain on the periphery, forming groups of their own. Squirrel monkeys in captivity have been known to live for up to twenty-five years.

### 2. The environment and its control

# 2.1. Ventilation

(See paragraph 2.1. of the General section)

# 2.2. Temperature

Though the species live in a wide range of climatic conditions in tropical forests from low to high altitudes in mountain areas, temperature changes in the habitats of individual colonies or troops do not vary greatly. Therefore marked short-term temperature variations should be avoided. In the wild the animals adapt to ambient temperatures by choosing the most suitable level within the canopy (for example, nearer to the ground in cool weather). Whereas normal room temperatures of 22  $^{\circ}$ C to 26  $^{\circ}$ C seem to be adequate, for animals with restricted exercise areas temperatures around 26  $^{\circ}$ C may be more appropriate.

# 2.3. Humidity

A range of 40 to 70 % is adequate for this species.

# 2.4. Lighting

As tropical-forest dwellers, squirrel monkeys are adapted to diffuse lighting. Nevertheless, for animals without access to outdoor enclosures, areas with high intensities of light similar to daylight should be provided. The light spectrum should resemble daylight even though the light intensity need not be that of bright sunshine. A 12 hour/12 hour light and dark cycle is appropriate. The daylight period should not be less than eight hours. The addition of a UV component or time-limited exposure to UV lamps would enable essential vitamin D3 synthesis in skin.

# 2.5. Noise

(See paragraph 2.5. of the General considerations for non-human primates)

### 2.6. Alarm systems

(See paragraph 2.6. of the General considerations for non-human primates)

### 3. Health

Squirrel monkeys may be silent carriers of a herpes virus (Saimirine herpesvirus 1, syn. Herpesvirus tamarinus, herpes T, Herpesvirus platyrrhinae), which, when transmitted to marmosets, may prove fatal. It is, therefore, recommended to not keep these two animal species in the same units unless tests have shown the colonies to be free from this viral infection.

# 4. Housing, enrichment and care

### 4.1. Housing

Based on their natural social organisation there is no difficulty in keeping saimiris in large single-sex groups. For this purpose, however, male and female groups should be well separated to avoid fighting. Special attention should be paid to identify distressed individuals in a group since aggressive behaviour is not very pronounced in squirrel monkeys.

For breeding purposes a group of seven to ten females kept with one or two males appears to be adequate. Breeding groups should have visual contact, but should be prevented from physical contact, with other groups.

Newborn animals are carried on the backs of their mothers until they are about 6 months old. However, they leave their mothers for exploration or are carried by close relatives at quite an early stage. They thus learn to socialise and, frequently through vocalisations, discover what may be dangerous or beneficial for them. The animals take up solid food from the age of three months onward. Nevertheless it is recommended that young animals should not be separated from their families before 6 months of age or, if hand feeding is necessary, they can be placed for adoption by another female, if possible, in their natal group. Squirrel monkeys reach sexual maturity at about the age of 3 years.

Breeding groups, once established, should not be disturbed, to avoid reduction in breeding performance. Major environmental and social changes should thus be avoided.

### 4.2. Enrichment

As arboreal animals, squirrel monkeys need sufficient climbing possibilities which can be provided by wire-mesh walls, poles, chains or ropes. Though they do leap over gaps if provided with structures, they prefer to run along or swing on horizontal and diagonal branches or rope bridges. Perches or nest boxes where they can sit huddled together for resting and sleep will be utilised.

A solid base with a substrate encourages foraging activity and play. The animals should be offered a choice of sites within the enclosure to allow for activity, to enable them to retreat from social contact and to allow them to select comfortable temperatures and lighting conditions.

# 4.3. Enclosures — dimensions and flooring

Table F.2.

Squirrel Monkeys: Minimum enclosure dimensions and space allowances

Minimum floor area for 1 (*) or 2 animals (m²)	Minimum volume per additional animal over 6 months of age (m³)	Minimum enclosure height (m)	
2,0	0,5	1,8	

<sup>(\*)</sup> Animals should only be kept singly under exceptional circumstances (see paragraph 4.1). Squirrel monkeys should preferably be kept in groups of 4 or more animals.

# 4.4. Feeding

Squirrel monkeys require a high protein intake. As with other South American species, squirrel monkeys require high levels of vitamin  $D_3$  in addition to vitamin C. Pregnant females are susceptible to folic acid deficiency, and should be provided with an appropriate powder or liquid supplement containing synthetic folic acid.

# 4.5. Watering

(See paragraph 4.7. of the General section)

# 4.6. Substrate, litter, bedding and nesting material

(See paragraph 4.6. of the General considerations for non-human primates)

# 4.7. Cleaning

(See paragraph 4.9. of the General section)

### 4.8. Handling

Squirrel monkeys can be trained to come forward for titbits or drinks as rewards. They are also capable of learning how to solve tasks for reward. For catching for investigation or treatment, animals should be trained to enter gangways with trap cages or individual enclosures.

# 4.9. Humane killing

(See paragraph 4.11. of the General section)

### 4.10. Records

(See paragraph 4.10. of the General considerations for non-human primates)

# 4.11. Identification

(See paragraph 4.11. of the General considerations for non-human primates)

### 5. Training of personnel

(See paragraph 5 of the General considerations for non-human primates)

# 6. Transport

(See paragraph 6 of the General considerations for non-human primates)

# d. Additional guidelines for housing and care of macaques and vervets

# 1. Introduction

The three species of macaque which are most commonly kept in laboratories all originate from Asia: Macaca mulatta (the rhesus monkey), Macaca fascicularis (the long-tailed, crab-eating or cynomolgus macaque) and Macaca arctoides (the stump-tailed or bear macaque). The vervet (Cercopithecus aethiops or Chlorocebus aethiops) is a rather similar type of African monkey sometimes kept in laboratories. In the wild, all of these species live in matriarchal multi-male/multi-female groups. There are both male and female dominance hierarchies and females form kinship groups within the troop. Social bonds are strongest between related females, and males compete for access to females in oestrus. Two species, the rhesus monkey and stump-tailed macaque live in warm to temperate climates, while the long-tailed macaque is an exclusively tropical species which particularly favours mangrove swamps and often forages in water. The long-tailed macaque is the most arboreal of the four species and the stump-tailed macaque the most terrestrial. The vervet has a wide range of African habitats, including open grasslands, forests and mountains, with climatic conditions ranging from warm temperate to tropical. Rhesus monkeys are seasonal breeders while the other species breed all year round in captivity. All the species have a predominantly vegetarian diet, although they may also feed on insects. Macaques and vervets in captivity have been known to live for more than thirty years.

### 2. The environment and its control

### 2.1. Ventilation

(See paragraph 2.1. of the General section)

# 2.2. Temperature

Rhesus monkeys and stump-tailed macaques are tolerant of temperate climates, vervets are also adaptable and temperatures of  $16~^{\circ}\text{C}$  to  $25~^{\circ}\text{C}$  are suitable. For the long tailed macaque, however, a more suitable range is  $21~^{\circ}\text{C}$  to  $28~^{\circ}\text{C}$ , although it will venture outdoors in much cooler weather.

### 2.3. Humidity

(See paragraph 2.3. of the General considerations for non-human primates)

# 2.4. Lighting

(See paragraph 2.4. of the General considerations for non-human primates)

### 2.5. Noise

(See paragraph 2.5. of the General considerations for non-human primates)

### 2.6. Alarm systems

(See paragraph 2.6. in the General considerations for non-human primates)

### 3. Health

Old World monkeys belong to the most susceptible species for tuberculosis and a high percentage of Asiatic macaques in the wild are silent carriers of Herpes B (syn. Herpes simiae, Cercopithicine herpesvirus 1). Vervets may also be susceptible to Marburg Virus and Ebola Virus.

### 4. Housing, enrichment and care

### 4.1. Housing

Macaques and vervets should be kept with social companions. Should larger groupings be feasible, this should be encouraged. Same-sex groups are most easily created at the time when the animals are separated from their mothers. With all social housing, staff should be vigilant to ensure that aggression is minimised. Vervet colonies are particularly prone to outbreaks of violence, especially after any form of disturbance to the group.

Breeding groups in captivity will usually be composed of one male and six to twelve females. With larger groups, to improve conception rates, two males can be included. If one male is considerably younger than the other, competition between them will be reduced. Where linked enclosures are used, care should be taken to monitor female-female aggression when the male is out of sight in the other part of the enclosure.

The age of removal of young macaques from their mothers is an important consideration for the breeding female, future breeders and stock animals. The young should not normally be separated from their mothers earlier than 8 months of age, preferably 12 months, apart from infants which are unable to be reared by their mother, for example due to poor lactation, injury or illness. To avoid major behavioural disturbances, such hand-reared animals should be re-integrated with other compatible animals as soon as possible. Separation before six months can cause distress and may lead to persistent behavioural and physiological abnormalities.

# 4.2. Enrichment

These animals, having advanced cognitive capabilities, require a suitably complex environment. A solid floor, which can be enriched by providing a non-toxic substrate, will allow for the concealment of scattered food items and encourage foraging. The enclosures should include vertical and diagonal structures for climbing, facilitating the use of the whole volume of the enclosure. Shelves and perches should not be placed one above the other. A space should be left between the shelf and enclosure wall to allow for the animal to suspend its tail freely.

Ladders, perches and toys to chew are all of value. In larger enclosures, a water tank (which is easily emptied) is particularly valuable for M. fascicularis but M. mulatta will also use it. Food can be dropped into the water for the long-tailed macaque and it will dive to retrieve it. Devices to encourage foraging (ranging from food scattered in the substrate to puzzle-feeders) have proved effective. Suitable food material can be placed on the mesh roof to encourage the animals to access it from the top of the enclosure. As novelty is important, toys should be provided and exchanged frequently.

### 4.3. Enclosures — dimensions and flooring

For the animals to feel secure, the design and interior dimensions of the enclosure should at least allow them to climb above human eye level.

Housing the animals in groups and in enclosures larger than the minimum group sizes and enclosure dimensions proposed in table F.3 should be encouraged

Table F.3. Macaques and vervets: Minimum enclosure dimensions and space allowances (\*)

	Minimum enclosure size (m²)	Minimum enclosure volume (m³)	Minimum volume per animal (m³)	Minimum enclosure height (m)
Animals less than 3 yrs of age (**)	2,0	3,6	1,0	1,8
Animals from 3 yrs of age (***)	2,0	3,6	1,8	1,8
Animals held for breeding purposes (****)			3,5	2,0

- Animals should only be kept singly under exceptional circumstances (see paragraph 4.1). An enclosure of minimum dimensions may hold up to three animals. An enclosure of minimum dimensions may hold up to two animals.
- In breeding colonies no additional space/volume allowance is required for young animals up to 2 years of age housed with their mother.

Animals should be housed in indoor enclosures providing appropriate environmental conditions of sufficient size to permit all animals to be provided with at least the minimum space allowances set out in table F.3 above.

In certain climates, it may be possible to hold breeding and stock animals in entirely outdoor enclosures if adequate shelter from climatic extremes is provided.

#### 4.4. Feeding

(See paragraph 4.4. in the General considerations for non-human primates)

#### 4.5. Watering

(See paragraph 4.7. of the General section)

#### 4.6. Substrate, litter, bedding and nesting material

(See paragraphs 4.3. and 4.6. of the General considerations for non-human primates)

#### 4.7. Cleaning

(See paragraph 4.9. of the General section)

#### 4.8. Handling

Macaques can easily be trained to cooperate in simple routine procedures such as injections or blood sampling and to come to an accessible part of the enclosure.

#### 4.9. Humane killing

(See paragraph 4.11. of the General section)

#### 4.10. Records

(See paragraph 4.10. of the General considerations for non-human primates)

#### 4.11. Identification

(See paragraph 4.11. of the General considerations for non-human primates)

#### 5. Training of personnel

(See paragraph 5 of the General considerations for non-human primates)

#### 6. **Transport**

(See paragraph 6 of the General considerations for non-human primates)

# e. Additional guidelines for housing and care of baboons

### 1. **Introduction**

Baboons include three genera, *Papio, Theropithecus* and *Mandrillus*, in which the commonly used species are *Papio papio* (Guinea baboon) and *Papio anubis* (Olive baboon).

Baboons inhabit woodlands and savannahs, including arid steppes and mountain deserts. They are heavily built terrestrial and quadrupedal animals. They display a great prognathism. Males are equipped with large canines.

Baboons are omnivorous and eat a wide variety of foods, mostly vegetarian (fruit and roots), although they do eat insects and occasionally mammal prey such as young gazelles or other non-human primates.

Papio papio and Papio anubis live in multi-male/multi-female groups.

Baboons in captivity have been known to live for more than thirty-five years.

The following guidelines are relevant to Papio papio and Papio anubis.

# 2. The environment and its control

### 2.1. Ventilation

(See paragraph 2.1. of the General section)

### 2.2. Temperature

Baboons are tolerant and adaptable of temperate climates and temperatures of 16 °C to 28 °C are suitable.

# 2.3. Humidity

(See paragraph 2.3. of the General considerations for non-human primates)

# 2.4. Lighting

(See paragraph 2.4. of the General considerations for non-human primates)

# 2.5. Noise

(See paragraph 2.5. of the General considerations for non-human primates)

# 2.6. Alarm system

(See paragraph 2.6. of the General considerations for non-human primates)

# Health

(See paragraph 3 of the General considerations for non-human primates)

# 4. Housing, enrichment and care

### 4.1. Housing

Adults and juveniles should be kept with social companions. Stock animals can be kept in compatible same-sex groups. Wherever possible, experimental animals should be kept in same-sex pairs or groups.

Breeding groups should be composed of one male and six to seven females, or two males and twelve to fifteen females. Larger groups may be much more difficult to manage. Staff should be vigilant to ensure that aggression is minimised. Baboon colonies are particularly prone to outbreaks of aggression, especially after any form of disturbance to the group.

The young should not normally be separated from their mothers before eight months of age, preferably twelve months, apart from infants which have been rejected or whose mother is not lactating adequately, or other veterinary reasons.

#### 4.2. Enrichment

Baboons, having advanced cognitive capabilities, require a suitably complex environment. A solid floor, which can be enriched by providing a non-toxic substrate, will allow for the concealment of scattered food items and encourage foraging. Ladders, perches and toys to chew are all of value. Food may be placed on the mesh roof to encourage the animals to access it from the top of the enclosure. Due to the size and the behavioural needs of baboons, enclosures should be robust and include broad shelves and blocks. As novelty is important, toys should be provided and exchanged frequently.

#### 4.3. Enclosure — dimensions and flooring:

For the animals to feel secure, the design and interior dimension of the enclosure should be at least high enough to allow them to climb above human eye level.

Housing the animals in groups and in enclosures larger than the minimum group sizes and enclosures dimensions proposed in table F.4 should be encouraged

Table F.4. Baboons: Minimum enclosure dimensions and space allowances (\*)

	Minimum enclosure size (m²)	Minimum enclosure volume (m³)	Minimum volume per animal (m³)	Minimum enclosure height (m)
Animals (**) less than 4 yrs of age	4,0	7,2	3,0	1,8
Animals (**) from 4 yrs of age	7,0	12,6	6,0	1,8
Animals held for breeding purposes (***)			12,0	2,0

Animals should only be kept singly under exceptional circumstances (see paragraph 4.1.). An enclosure of minimum dimensions may hold up to 2 animals.

Animals should be housed in indoor enclosures providing appropriate environmental conditions of sufficient size to permit all animals to be provided with at least the minimum space allowances set out in table F.4. above.

In certain climates, it may be possible to hold breeding and stock animals in entirely outdoor enclosures if adequate shelter from climatic extremes is provided.

Enclosures should have a solid floor.

#### 4.4. Feeding

(See paragraph 4.4. of the General considerations for non-human primates)

#### 4.5. Watering

(See paragraph 4.7. of the General section)

#### 4.6. Substrate, litter, bedding and nesting material

(See paragraphs 4.3. and 4.6. of the General considerations for non-human primates)

#### 4.7. Cleaning

(See paragraph 4.9. of the General section)

#### 4.8. Handling

Baboons can be easily trained to cooperate in simple routine procedures such as injections or blood sampling and to come to an accessible part of the enclosure. However, for personnel safety considerations, great care should be taken in handling adult animals and suitable restraint deployed.

In breeding colonies no additional space/volume allowance is required for young animals up to 2 years of age housed with their mother.

# 4.9. Humane killing

(See paragraph 4.11. of the General section)

### 4.10. Records

(See paragraph 4.10. of the General considerations for non-human primates)

### 4.11. Identification

(See paragraph 4.11. of the General considerations for non-human primates)

# 5. Training of personnel

(See paragraph 5 of the General considerations for non-human primates)

# 6. Transport

(See paragraph 6 of the General considerations for non-human primates)

### G. SPECIES-SPECIFIC GUIDELINES FOR FARM ANIMALS AND MINI-PIGS

### a. General considerations

### 1. **Introduction**

For the purposes of these guidelines the term 'farm animals' includes cattle, sheep, goats, pigs, mini-pigs and equines, including horses, ponies, donkeys and mules.

The use of farm animals in research varies from applied experiments under farm conditions to more fundamental studies in agricultural, veterinary or biomedical research carried out under laboratory conditions. In the former case, it is important that the housing and management conditions, whilst taking due account of animal health and welfare, produce information which can be reliably applied to commercial farm conditions. In the latter case, where more invasive procedures are frequently involved, a different type of housing and management is necessary. The precise nature of the housing adopted should be suitable to yield information of relevance to the experimental question and appropriate for the procedures involved.

Management systems for all farm animals should accommodate their natural behaviour, in particular the need to graze or forage, exercise and socialise. Farm animals are held in a number of different types of enclosures, often dependent on experimental requirements. For example, farm animals may be held on pasture, in open-sided buildings with access to open yards, in enclosed buildings with natural ventilation or in specialised buildings for quarantine and biocontainment with natural or forced ventilation.

During agricultural research, when the aim of the research requires that the animals are kept under similar conditions to those under which commercial farm animals are kept, the keeping of the animals should at least follow the standards laid down in Council Directive 98/58/EC (²) and in the specific Directives for the protection of calves and pigs (Council Directives 91/629/EEC (³) and 91/630/EEC (⁴) as well as in the Recommendations adopted under the Council of Europe Convention for the Protection of Animals kept for Farming Purposes (ETS No 87).

### 2. The environment and its control

Under natural conditions farm animals are exposed to, and will tolerate, a wide range of temperatures, although there is some variation in the degree of tolerance between species and breeds. They will seek shelter against driving rain and strong wind, and protection from intense sun. Where they are kept in enclosures exposed to outdoor conditions, shelter and shade and a reasonably dry lying area should be provided. Shelters should be carefully positioned taking these factors into consideration. Sufficient shelter should be provided to protect all animals from adverse climatic conditions.

<sup>(2)</sup> OJ L 221, 8.8.1999, p. 23.

<sup>(3)</sup> OJ L 340, 11.12.1991, p. 28.

<sup>(4)</sup> OJ L 340, 11.12.1991, p. 33.

Animals held outdoors or in buildings with natural ventilation will be exposed to ambient environmental conditions. Animals should not be restricted to such areas under climatic conditions which may cause the animals distress.

Environmental parameters, in particular temperature and humidity, are strictly interrelated and should not be considered in isolation.

### 2.1. Ventilation

All farm animals are sensitive to respiratory problems. In the absence of mechanical ventilation, as is the case in a significant number of farm animal buildings, it is important to ensure that suitable air quality is provided by natural ventilation (see paragraph 2.1.1. of the General section).

Dust levels in the air from feed and bedding should be minimised.

# 2.2. Temperature

The thermoneutral zones of farm species vary considerably, depending on the conditions to which the animals are acclimatised. Farm animals living outdoors develop a thick layer of hair/wool during the winter months to help them to tolerate low temperatures. They may acclimatise to lower temperatures indoors even without the growth of winter coats, provided the relative humidity is low, draughts are avoided and they have a lying area with sufficient bedding material. In indoor enclosures it is therefore important to avoid wide fluctuations and sudden changes in temperature, particularly when moving animals between indoor and outdoor accommodation. As farm animals may suffer from heat stress, during periods of high temperature it is important to ensure that appropriate measures, for example the shearing of sheep and provision of shaded lying areas, are in place to avoid welfare problems.

Appropriate temperature ranges are dependent on a number of factors including, for example, breed, age, caloric intake, weight, stage of lactation and type of environment.

# 2.3. Humidity

Under natural conditions, farm animals are exposed to, and tolerate well, a wide range of relative humidities. In controlled environments extremes and sudden wide fluctuations of humidity should be avoided, as both high and low humidity can predispose animals to disease.

In indoor enclosures, buildings should be designed with sufficient ventilation to prevent prolonged periods of high humidity, as this may cause excessive dampness in the animal enclosures, predisposing the animals to respiratory disease, foot-rot and other infectious conditions.

# 2.4. Lighting

Farm species have evolved to live in different conditions; for example ruminants graze and rest during daylight in open grassland, whereas pigs show crepuscular activity in woodland areas. Provision of adequate light is important for all farm animal species, and natural light is preferred where possible. Where this is not provided, the light part of the photoperiod should be within a range of eight to twelve hours daily, or should reproduce natural light cycles. A controlled photoperiod may be needed for breeding and for some experimental procedures. Sufficient natural or artificial light should also be available for inspection of groups and individuals.

Where windows are provided, breakable glass should be screened using a protective physical barrier or be situated out of reach of the animals.

### 2.5. Noise

Unavoidable background noise from, for example, ventilation equipment, should be minimised, and sudden noises should be avoided. Handling and restraint facilities should be designed and operated to minimise noise during use.

# 2.6. Alarm systems

(See paragraph 2.6. of the General section)

### 3. Health

### 3.1. Disease control

As farm animals are often sourced from commercial farms, it is important that measures are taken to ensure that animals of a suitable health status are obtained. Mixing animals from different sources is a particular risk.

Preventive medicine programmes should be developed on the basis of veterinary advice for all farm species, and appropriate vaccination regimes adopted as necessary.

Foot care management, parasite control measures and nutritional management are essential parts of all farmanimal health programmes. Regular dental examinations and respiratory disease preventive measures are of particular importance in equine programmes.

Regular review of production indices and condition scoring should also be included.

Care is needed to ensure that any substrate provided does not introduce or promote growth of infectious agents or parasites.

### 3.2. Behavioural abnormalities

Behavioural abnormalities such as tail, ear or flank chewing or biting, wool pulling, navel sucking, weaving and crib biting can occur as a consequence of poor husbandry or environmental conditions, social isolation, or from boredom due to long periods of inactivity. If such abnormalities occur, measures should be taken immediately to rectify these deficiencies including, for example, a review of environmental factors and management practices.

# 3.3. Husbandry

Disbudding, dehorning of adult animals, castration and tail docking should not be done unless justified on welfare or veterinary grounds. When those techniques are carried out, appropriate anaesthesia and analgesia should be provided.

# 3.4. Neonatal care

High standards of stockmanship and care are necessary for successful rearing of farm animals during the neonatal period.

Suitable accommodation, with a dry clean area, should be provided for peri-parturient and neonatal animals. Facilities should be designed to facilitate observation and be maintained to high hygiene standards, as young animals are particularly susceptible to infections.

All neonates should receive adequate amounts of colostrum as soon as possible after birth, and preferably within four hours. Adequate supplies of colostrum should be available for use in emergencies.

Suitable feeding practices should be in place to allow normal growth and development, with access to roughage provided to ruminants from two weeks of age.

As neonatal animals have poor thermo-regulatory control, particular care is needed to ensure that suitable temperatures are provided and maintained. A supplementary local heat source may be required, although care is needed to avoid the risk of injury, such as burns, and accidental fires.

To reduce the risk of mis-mothering or rejection, it is important that a strong maternal bond is allowed to develop during the first few days of life. During this period it is important to minimise handling or management procedures, such as transport, castration or tagging, that may disrupt this relationship or prevent the young animals accessing sufficient amounts of colostrum or milk.

Weaning strategies should be given due consideration to minimise stress in the mother and offspring. Weaning into groups of animals of similar ages facilitates the development of compatible and stable social structures.

Naturally reared pigs and mini-pigs should not be weaned before four weeks of age, lambs, kids and beef calves before six weeks of age and equines before twenty weeks of age, unless there is justification on veterinary or welfare grounds.

For animals which are artificially reared, commonly dairy calves, appropriate feeding regimes should be provided to satisfy nutritional requirements, and in the case of ruminants, to promote normal rumen development.

Early weaning from the dam on experimental or veterinary grounds should be determined in consultation with the animal technician and with the competent person charged with advisory duties in relation to the well-being of the animals. In such circumstances, additional attention and means should be targeted to the welfare and care of these animals.

# 4. Housing, enrichment and care

# 4.1. Housing

Farm animals should be housed in socially harmonious groups within the animal enclosure, and husbandry practices designed to minimise social disruption, unless the scientific procedures or welfare requirements make this impossible.

When kept in groups, a defined hierarchy is quickly established. Some aggressive interaction may be encountered during initial grouping while relative rankings in the social hierarchy are established.

Special care is needed to minimise aggression and potential injury when grouping, regrouping, or introducing an unfamiliar animal to a group. In all cases, animals should be grouped according to size and age and monitored for social compatibility on an ongoing basis.

Separation from a group, and the single-housing of farm animals for even short periods can be a significant stress factor. Therefore, farm animals should not be single-housed unless justified on welfare or veterinary grounds. The exceptions, where animals may prefer to be housed singly include females about to give birth, and adult boars, which can be solitary under natural conditions.

Single-housing on experimental grounds should be determined in consultation with the animal technician and with the competent person charged with advisory duties in relation to the well-being of the animals. Factors to be taken into consideration should include the nature of the individual animals, their likely reaction to separation from the group and the need for and duration of an habituation period. Where individual housing is necessary, animals should have visual, auditory and olfactory contact with conspecifics.

### 4.2. Enrichment

As a stimulating environment is an important contributing factor to farm animal welfare, environmental enrichment should be provided to prevent boredom and stereotypic behaviour. All farm animal species naturally spend a large amount of time each day grazing, browsing or rooting for food, and in social interaction. Suitable opportunities should be provided to meet these behaviours, by for example access to pasture, the provision of hay or straw or manipulable objects such as chains or balls.

Enrichment materials and devices should be changed at regular intervals since animals, in particular pigs, tend to lose interest in materials to which they have become accustomed. Sufficient enrichment devices should be provided to minimise aggressive behaviour

### 4.3. Enclosures — dimensions and flooring

Appropriate design of farm-animal enclosures is essential to ensure that suitable space is available within the enclosure to allow the animals to carry out a range of normal behaviour. Floor type, drainage, provision of bedding (and hence ease of maintaining hygiene) and the social circumstances (group size and stability) will all impact on the space requirements for the animals.

All enclosures should be designed and maintained to ensure that animals cannot be trapped or injured, for example in partitions or under feeding troughs.

Animals should not be tethered, unless justified on scientific or veterinary grounds, in which case this should be for the minimum time period necessary.

Sufficient space should be provided for each animal to stand up, lie comfortably, stretch and groom themselves, with access to a communal lying area and adequate room for feeding.

The lying area should allow all animals to lie in lateral recumbency simultaneously, bearing in mind that whilst some farm animals, for example pigs, generally prefer to lie in physical contact with other conspecifics, others, such as equines prefer a degree of spatial separation. Under conditions of high temperatures, where animals need to lie with complete spatial separation to facilitate heat loss, a greater lying area should be allowed.

The lying area should be provided with bedding to enhance comfort and reduce the incidence of pressure lesions. Where absence of bedding is necessary for experimental reasons, the floor should be designed and insulated to improve physical and, unless a suitable controlled environment is provided, thermal comfort.

The height of enclosures should allow natural rearing and mounting behaviour.

Enclosure flooring materials should be non-injurious and provide adequate grip for unconstrained locomotion and posture change. Floors should be well maintained and replaced when necessary, as surface damage causing injuries will develop over time.

### 4.4. Feeding

The diet should provide adequate nutrients to support the maintenance energy requirements of each animal, given the environmental conditions under which animals are kept. Additional energy will be needed to support pregnancy, lactation and growth, and should be tailored to the needs of the animals (for example, high genetic merit dairy cattle). Vitamin and mineral levels in the diet should also be considered, for example to avoid copper toxicity in sheep or the formation of urinary calculi in male castrated sheep, and where necessary, mineral licks should be provided.

When grazed grass is used as forage, stocking densities should be controlled to ensure adequate supplies are available to meet the nutritional requirements of all the animals. Where grass supply is limited, provision of additional feed in the field should be considered.

For ruminants and horses, sudden changes in diet should be avoided, and new items introduced gradually, especially where high-energy feeds are introduced, or during periods of high metabolic demand, for example around parturition. Sufficient roughage should be provided.

In group-housing systems, there should be sufficient food provided in sufficient numbers of sites for all individuals to access without risk of injury.

Forage forms a significant component of the diets of farm animals. Since the amount of forage needed may preclude the use of bags for storage, forage items, including hay, straw, silage and root crops, should be stored in a way that minimises deterioration in quality and the risk of contamination. A pest-control strategy should be in place in areas where forage and concentrates are stored.

When grass is cut for feeding housed animals (for example, zero-grazing), it should be done frequently, as cut grass heats up when stored and becomes unpalatable.

### 4.5. Watering

Animals should have access at all times to fresh uncontaminated water, which should be readily accessible to all individuals within the social group. The number of drinking points or trough length should be sufficient to allow access to water for all individuals within the social group. Flow rates should meet the demands of the individual animal as these will vary depending on the feed, physiological status and ambient temperature, for example, lactating animals have much higher water demands than stock animals.

# 4.6. Substrate, litter, bedding and nesting material

(See paragraph 4.8. of the General section)

# 4.7. Cleaning

(See paragraph 4.9. of the General section)

### 4.8. Handling

If handling and restraint facilities are required, these should be of robust construction and safe for animals and operators. In particular, a non-slip floor should be provided.

Handling and restraint facilities can take the form of basic equipment provided within the animal enclosure, or more complex, dedicated facilities serving the needs of the whole establishment. Handling and restraint facilities can be provided in the enclosure area, but care should be taken to ensure that these do not compromise space allowances or create a potentially hazardous physical obstruction in the enclosure.

The dedicated facilities should, where possible, incorporate races and pens for separating animals; footbaths; special facilities for some species such as plunge dip baths and shearing pens for sheep; and an area to allow animals to recover after treatments. Ideally these facilities should be protected from prevailing weather conditions for the comfort of both animals and operators.

Animals should be handled quietly and firmly and not be rushed along races and passageways. These should be designed, taking account of the natural behaviour of the animals, to facilitate ease of movement and minimise the risk of injury. Immobilisation devices should not cause injury or unnecessary distress. Aversive stimuli, physical or electrical, should not be used.

Passages and gates should be of sufficient width to permit two animals to pass freely, whereas races should be of width only to permit one-way movement.

Regular handling will allow habituation of animals to human contact. Where frequent handling is required, a programme of training and positive rewards should be considered to minimise fear and distress.

Animals should not be closely confined except for the duration of any examination, treatment or sampling, whilst accommodation is being cleaned, collecting for milking, or loading for transport.

# 4.9. Humane killing

All systems for the humane killing of farm animals should be designed to ensure that animals are not caused unnecessary distress. Careful handling by experienced staff, with minimum disruption to normal practices, will minimise distress to the animals, before they are humanely killed.

Killing should not be performed in areas where other animals are present, unless in the case of euthanasia of a badly injured animal where additional suffering may be caused by moving the animal.

# 4.10. Records

(See paragraph 4.12. of the General section)

### 4.11. Identification

Animals should be individually identified by the appropriate use of transponders, ear tags, plastic neck collars and/or rumen boluses. Freeze branding and tattooing may be less suitable. Hot branding should not be used.

Identification devices should only be applied by trained personnel and at times when the procedure is likely to have minimal adverse effects on the animal. Tagged or tattooed ears should be checked regularly for signs of infection and lost tags should be replaced using the original tag hole where possible.

If electronic devices of identification are used, they should be of the correct size and specification for the animal and should be checked regularly for function and the absence of any adverse reactions, for example, injection site reactions and rubbing or pharyngeal trauma as a result of improper bolus administration.

# b. Additional guidelines for housing and care of cattle

### 1. **Introduction**

Cattle (Bos taurus and Bos indicus) are social animals forming hierarchies based on dominance relationships among herd members. They will frequently develop affinity relationships with conspecifics. As ruminants, cattle spend much of the day foraging, followed by long rest periods. Cattle are normally docile and are easily habituated to human contact.

### 2. The environment and its control

(See paragraph 2 of the General considerations for farm animals and mini-pigs)

### 3. Health

(See paragraph 3 of the General considerations for farm animals and mini-pigs)

# 4. Housing, enrichment and care

### 4.1. Housing

Horned and polled animals should not be mixed, except for young calves and their mothers.

### 4.2. Enclosures — dimensions and flooring

 $\label{eq:continuous} \textit{Table G.1.}$  Cattle: Minimum enclosure dimensions and space allowances

Body weight (kg)	Minimum enclosure size (m²)	Minimum floor area/animal (m²/animal)	Trough space for ad-libitum feeding of polled cattle (m/animal)	Trough space for restricted feeding of polled cattle (m/animal)
Up to 100	2,50	2,30	0,10	0,30
over 100 to 200	4,25	3,40	0,15	0,50
over 200 to 400	6,00	4,80	0,18	0,60
over 400 to 600	9,00	7,50	0,21	0,70
over 600 to 800	11,00	8,75	0,24	0,80
over 800	16,00	10,00	0,30	1,00

Where cattle are housed indoors, a bedded area sufficient to allow all of the animals to lie simultaneously will be provided. Where cubicles are not provided, this area will normally be approximately 70 % of the minimum floor area shown in the above table. The remainder of the enclosure can be non-bedded for feeding and exercise.

If individual open-ended cubicles are provided as the bedded area, this area may be reduced in size, but the total number of cubicles should exceed animal numbers by 5 % to reduce competition and permit all animals to lie simultaneously. The design of cubicles is critical to their comfort, and specialist advice should be sought before installation. It should include consideration of the body size of the animal, a surface sufficiently cushioned to prevent injury, adequate stall drainage, correctly positioned stall dividers and head rails, lateral and vertical freedom for head movement and adequate lunging space. The height of the rear step should prevent dung entering the cubicle during cleaning, but not be of such a height that it causes damage to the feet during ingress and exit. The remainder of the enclosure can be non-bedded for feeding and exercise.

Cubicle length is primarily determined by the weight of the animals. Cubicle width will vary, depending on the type of division used, but must be sufficient to allow the animals to lie comfortably without undue pressure being exerted by the divisions on vulnerable parts of the body. Specialist advice should be sought on the design and installation of cubicles.

# 4.3. Feeding

The trough space provided will allow all animals to feed at the same time, unless the diet is available ad libitum (see above table). Horned cattle require more trough space than polled animals, and due allowance should be made for this.

# 4.4. Watering

Water troughs: there should be sufficient linear trough space to allow 10 % of the animals to drink at one time. This equates to a minimum of 0,3 metres per 10 adult cattle. Lactating dairy cows will require 50 % more space.

Water bowls: a minimum of two water bowls should be provided when cattle are group-housed. For groups of over twenty cattle, at least one drinking bowl for ten animals should be provided.

### 4.5. Handling

Where animals are milked by machine, equipment should be maintained to a high standard to prevent diseases such as mastitis.

Horned cattle may present a danger to personnel in confined spaces. Under these circumstances, it may be necessary to consider dehorning. Wherever possible, this should be carried out on calves under the age of eight weeks.

### c. Additional guidelines for housing and care of sheep and goats

### 1. Introduction

Sheep (Ovis aries) are grazing animals which, because of differences between breeds, for example fleece characteristics, will thrive in a wide range of climatic conditions.

Under natural or farming conditions, sheep are very social, spending all their lives close to other members of the flock whom they recognise individually. As a species, they are therefore particularly disturbed by social isolation, a factor which should be taken into account when designing animal accommodation. However, in terms of social cohesion there are recognisable variations between breeds as, for example, hill sheep tend not to flock closely together when left undisturbed.

Goats (*Capra hircus*) are a naturally inquisitive species and generally interact well with other animal species and humans. Like sheep, goats live in social groups and are disturbed by social isolation. Goats obtain their food by browsing more than by grazing and are best adapted to dry, firm ground. Their ability to climb is considerable and this facilitates their browsing. They prefer warm conditions and do not tolerate wet and windy conditions well.

# 2. The environment and its control

Under extreme conditions, sheep will require access to natural or artificial wind-break shelter and shade, whilst different coat characteristics mean that goats are less tolerant of prolonged rain and should have free access to roofed shelter areas whilst outside.

Recently shorn animals may need higher environmental temperatures than fleeced animals.

### 3. Health

Adult sheep and goats of wool breeds should be shorn at least once per year, unless this would compromise their welfare.

# 4. Housing, enrichment and care

# 4.1. Housing

Entire adult males from both species can be more solitary than females and young offspring. They may be aggressive, particularly during the breeding season, requiring careful management to reduce the risks of fighting and injury to handlers.

Horned and polled goats should not be housed together.

### 4.2. Enrichment

Sufficient raised areas of appropriate size and quantity to prevent dominant animals impeding access should be provided for goats.

### 4.3. Enclosures — dimensions and flooring

Table G.2. Sheep and Goats: Minimum enclosure dimensions and space allowances

Body weight (kg)	Minimum enclosure size (m²)	Minimum floor area/animal (m²/animal)	Minimum partition height (*) (m)	Trough space for ad-libitum feeding (m/animal)	Trough space for restricted feeding (m/animal)
Up to 20	1,0	0,7	1,0	0,10	0,25
over 20 to 35	1,5	1,0	1,2	0,10	0,30
over 35 to 60	2,0	1,5	1,2	0,12	0,40
over 60	3,0	1,8	1,5	0,12	0,50

<sup>(\*)</sup> For adult goats, an increased minimum partition height may be required to prevent escape.

The entire enclosure should have a solid floor with appropriate bedding provided.

### 4.4. Watering

In indoor enclosures for sheep and goats at least one drinking point per twenty animals should be provided.

### 4.5. Identification

Dyeing the fleece or coat using recognised non-toxic agricultural marker products may be used for short-term experiments in short-wool breeds of sheep and in goats.

# d. Additional guidelines for housing and care of pigs and mini-pigs

# 1. Introduction

The domestic pig (Sus scrofa) is descended from the European wild boar. Although subject to intensive selection pressure over many generations for production characteristics of economic importance, domesticated pigs have largely retained the same behavioural repertoire as their ancestors. Under unrestricted conditions, they live in small family groups, show a crepuscular diurnal rhythm and have strongly developed exploratory behaviour. They are omnivorous and a large part of their active time is spent foraging for food. At birth, sows farrow in social isolation and construct a nest prior to parturition. Weaning is gradual and is completed at about four months of age, and piglets integrate gradually into the social group with little aggression.

Mini-pigs differ from the farm pig in many significant respects. A number of different mini-pig strains have been developed by conventional breeding procedures in order to produce a small pig as a laboratory animal suitable for research purposes. For the purpose of this annex, the mini-pig is considered as a small pig breed for usage for experimental and other scientific purposes and with an adult body weight typically not exceeding 60 kg, but can be as high as 150 kg in some strains. Because of this difference in body size at maturity, recommendations for farm pigs cannot always be extrapolated on a simple weight basis. Recommendations in this document apply to both types of pig, with specific requirements of mini-pigs annotated where necessary.

# 2. The environment and its control

### 2.1. Temperature

Pigs and mini-pigs are highly sensitive to environmental temperature and place a high behavioural priority on thermoregulation.

Pigs may be kept in a uniform, temperature-controlled environment, in which case the whole room should be maintained within the thermoneutral zone. Alternatively, they may be kept in an enclosure with different microclimates, by providing localised heating or kennelling of the lying area and provision of adequate bedding material. A temperature gradient within the enclosure is considered beneficial. Outdoor pigs can compensate for lower ambient temperature provided that adequate shelter, with plentiful dry bedding, and additional food is provided.

Table G.3.

Pigs and minipigs: Guideline temperature ranges for single-housed animals

Liveweight	Recommended temperature range (°C)
Less than 3 kg	30 to 36
from 3 to 8 kg	26 to 30
over 8 to 30 kg	22 to 26
over 30 to 100 kg	18 to 22
over 100 kg	15 to 20

In addition to body weight, suitable temperatures will vary according to sexual maturity, the presence or absence of bedding, group housing, and the caloric intake of the animal. Within the ranges given, animals of lower body weight, without bedding or with restricted caloric intake should be provided with the higher temperatures.

Piglets of low body weight are very sensitive to environmental temperature and should be provided with higher temperatures. Litters of newborn piglets should be offered a lying area minimum of 30  $^{\circ}$ C, decreasing to 26  $^{\circ}$ C at the age of two weeks. For farrowing/lactation rooms, the minimum room temperature necessary is that required to allow an adequate temperature to be maintained in the piglet lying area, taking account of any local heat supply. Because of their high metabolic activity, lactating sows are prone to heat stress and farrowing room temperatures should ideally not exceed 24  $^{\circ}$ C.

# 3. Health

(See paragraph 3 of the general considerations for farm animals and mini-pigs)

# 4. Housing, enrichment and care

# 4.1. Enrichment

Pigs show spatial separation of different behaviours such as lying, feeding and excretion. Enclosures should therefore allow for the establishment of separate functional areas by providing either plentiful space or appropriate subdivision of the enclosure area.

Pigs have a high motivation to explore and should be provided with an environment of sufficient complexity to allow expression of species-specific exploratory behaviour. All pigs should at all times have access to adequate amounts of materials for investigation and manipulation, including rooting, in order to reduce the risk of behavioural disorders.

# 4.2. Enclosures — dimensions and flooring

Table G.4. indicates the minimum space requirement for an animal at any given liveweight. Enclosures should be designed to accommodate the highest liveweight that pigs will finally reach in any given circumstance. The number of enclosure changes should be minimised.

Table G.4.

Pigs and Minipigs: Minimum enclosure dimensions and space allowances

Liveweight (kg)	Minimum enclosure size (*) (m²)	Minimum floor area per animal (m²/animal)	Minimum lying space per animal (in, thermoneutral conditions) (m²/animal)
Up to 5	2,0	0,20	0,10
over 5 to 10	2,0	0,25	0,11
over 10 to 20	2,0	0,35	0,18
over 20 to 30	2,0	0,50	0,24
over 30 to 50	2,0	0,70	0,33
over 50 to 70	3,0	0,80	0,41
over 70 to 100	3,0	1,00	0,53
over 100 to 150	4,0	1,35	0,70
over 150	5,0	2,50	0,95
Adult (conventional) boars	7,5		1,30

<sup>(\*)</sup> Pigs may be confined in smaller enclosures for short periods of time, for example by partitioning the main enclosure using dividers, when justified on veterinary or experimental grounds, for example where individual food consumption is required.

Where pigs are housed individually or in small groups, greater space allowances per animal are required than for those in larger groups.

Pigs should not be tethered at any time, and should not be confined in stalls or crates except for short periods of time necessary for feeding, insemination, veterinary or experimental purposes. The accommodation for sows and piglets should enable the fulfilment of the special behaviour patterns of the sow before and after parturition, and those of the piglets after birth. Thus, although the use of farrowing crates can safeguard piglet survival and welfare under some conditions, the close confinement of sows during the perinatal and suckling periods should be limited as far as possible and loose housing systems should be aimed at.

The most appropriate flooring material will depend on the size and weight of the pigs. To facilitate provision of rooting/nesting substrate, it is desirable to provide a solid floor in the lying area of the pen. Slatted floors can be of value in facilitating good hygiene, but the slat and void dimensions should be appropriate to the size of the pigs in order to prevent foot injuries.

# 4.3. Feeding

Pigs kept for meat production are typically fed ad libitum until approaching maturity, after which restricted feeding practices are necessary to avoid obesity. Mini-pigs are prone to become obese on conventional pig diets. Special reduced calorie diets with increased fibre content help to prevent this problem. Where feed restriction is necessary, pigs will show increased foraging motivation which can be expressed as increased activity and aggression, and development of stereotyped oral behaviours. To avoid these problems it is important to modify diets to enhance satiety, for example by provision of increased dietary fibre, and to provide an appropriate foraging substrate such as straw.

With restricted feeding practices, young growing animals should be fed at least twice daily, whereas mature animals should be fed once daily, as an adequate meal size is important for the animal to reach satiety, and will minimise aggression. Where feeding is restricted, all individuals within the social group should have access to feed without causing aggression. Adequate trough space should be provided to ensure that animals can feed simultaneously. Recommended requirements are given in Table G.5. Where animals are housed singly or in small groups, the minimum trough space should be that for restricted feeding. When animals are housed in larger groups and fed ad libitum, trough space can be shared and a lower total space is required.

Table G.5.

Pigs and minipigs: Minimum feeding trough space allowances

Liveweight (kg)	Minimum trough space (cm) (ad-libitum and restricted feed- ing (*))	Minimum trough space per animal on ad-libitum feeding (cm/animal)
Up to 10	13	2,0
Over 10 to 20	16	2,5
Over 20 to 30	18	3,0
Over 30 to 50	22	3,5
Over 50 to 70	24	4,0
Over 70 to 100	27	4,5
Over 100 to 150	31	5,0
Over 150	40	7,0

<sup>(\*)</sup> Each animal on restricted feeding should be provided with at least the minimum trough space allowance.

# 4.4. Watering

As pigs are particularly sensitive to the consequences of water deprivation, in cases where they are group-housed, at least two drinking points per unit — or a large bowl allowing more than one pig to drink at the same time — should be provided to prevent dominant animals impeding access to the drinking point. To achieve this, the following drinking space allowances are recommended.

Table G.6.

Pigs and minipigs: Minimum drinking point allowances

Drinker type	No. of pigs per drinking point
Nipple or bite drinkers	10
Large bowl drinkers (which allow at least two pigs to drink at the same time)	20

Where pigs housed in larger groups are watered from an open trough, the minimum length of trough perimeter with access to water should be that allowing a single pig unimpeded access (as indicated in Table G.5. for restricted feeding space), or 12,5 mm of trough length per pig, whichever is the greater.

Table G.7.

Pigs and minipigs: minimum drinking water flow rates for pigs

Type of pig	Minimum water flow rate (ml/min)
Weaners	500
Growers	700
Dry sows and boars	1 000
Lactating sows	1 500

# 4.5. Substrate, litter, bedding and nesting material

Bedding contributes to pig welfare in many ways. It enhances physical and thermal comfort (except in hot environmental conditions), can be eaten to provide gut fill and enhance satiety, and provides a substrate for foraging and nest-building behaviours. The extent to which each of these different benefits can be provided will depend on the nature of the bedding, with long straw providing the best overall material but alternatives such as

chopped straw, sawdust, wood shavings and shredded paper conferring some benefits. Bedding should be non-toxic and, where possible, provide structural diversity to stimulate exploratory behaviour. Bedding should be provided for all pigs, unless precluded for experimental reasons, and is particularly important for farrowing sows, which have a strong motivation to perform nest-building behaviour, and for pigs on restricted feeding regimes, which have a strong motivation to express foraging behaviour.

# e. Additional guidelines for housing and care of equines, including horses, ponies, donkeys and mules

### 1. **Introduction**

Equines evolved as grazers of open grasslands, and domestic horses and ponies (*Equus caballus*) and donkeys (*Equus asinus*) have retained the behavioural repertoire of their ancestors. In the feral or free-ranging state, equines live in herds separated into small family groups or bands typically comprising one stallion, with several mares, foals and yearlings. The social structure develops as a clearly defined hierarchy, and individual animals within a group often form close pair bonds which it is important to recognise and maintain if possible. Mutual body care is a particularly important element in their social life.

Unlike ruminants, equines may graze continuously for many hours and under natural conditions they will spend fourteen to sixteen hours daily at this activity. Although their natural food is grass, herbs, and leaves, they are very selective regarding their choice of grass species and which part of the plant to eat. Their normal daily pattern is to graze, move a few steps and graze again. In this way they exercise as well as feed, and can cover long distances in a twenty-four hour period.

Ideally, management systems for equines should accommodate their natural behaviour, in particular the need to graze, exercise, and socialise. They are flight animals and hence easily startled and this should also be taken into account.

# 2. The environment and its control

# 2.1. Temperature

Rugs can be used in cool conditions, especially if hair has been clipped, but these should be removed and checked daily.

The mane and tail of equines provide protection from adverse weather conditions and from flies and should not be removed or cut short. Where manes and tails need to be shortened or tidied this should be achieved by trimming rather than by pulling.

### 3. Health

(See paragraph 3 of the General considerations for farm animals and mini-pigs)

# 4. Housing, enrichment and care

# 4.1. Enclosures — dimensions and flooring

Ideally, equines should be kept at pasture or have access to pasture for at least six hours a day. Where equines are kept with minimal or no access to grazing then additional roughage should be provided to extend the time spent feeding and reduce boredom.

In indoor enclosures, group-housing systems are preferred since these provide opportunities for socialisation and exercise. For horses it is essential that great care is taken to ensure social compatibility of groups

The total space requirement for indoor enclosures will depend on whether animals also have daily access to additional areas for grazing and/or other forms of exercise. The figures below assume that such additional areas will be provided. If not, then space allowances should be increased significantly.

# Table G.8. Equines: Minimum enclosure dimensions and space allowances

	Mi	Minimum floor area/animal (m²/animal)		
Wither height (m)	For each animal held singly or in groups of up to 3 animals	For each animal held in groups of 4 or more animals	Foaling box/mare with foal	Minimum enclosure height (m)
1,00 to 1,40	9,0	6,0	16	3,00
over 1,40 to 1,60	12,0	9,0	20	3,00
over 1,60	16,0	(2 × WH) <sup>2</sup> (*)	20	3,00

<sup>(\*)</sup> To ensure adequate space is provided, space allowances for each individual animal should be based on height to withers (WH).

The shortest side should be a minimum of  $1.5 \times$  the wither height of the animal.

The height of indoor enclosures should allow animals to rear to their full height to safeguard the welfare of the animals.

Slatted floors should not be used for equines.

### 4.2. Feeding

Incorrect feeding of equines can have very serious welfare implications, causing illnesses such as colic and laminitis.

Since they naturally graze for long periods, they should ideally have constant access to forage in the form of fresh grass, hay, silage or straw. Where they are not given the opportunity to graze, they should be provided with a suitable quantity of long fibre/roughage every day. Where possible roughage should be fed on the ground or in suitably designed round bale feeders. Hay nets and racks should be designed and positioned to minimise risk of injury.

If 'hard' (concentrate) feed is offered to animals, particularly where the animals are housed in groups the feeding order should, where possible, follow the herd order of dominance. Where possible individuals should be fed separately. If this is not possible feeding points should be spaced at least 2,4 m apart and there should be at least one point per animal. Horses fed with concentrates need to be given small amounts of feed frequently.

# 4.3. Watering

Horses prefer to drink from an open water surface, and this should be provided where possible. If automatic water nipple drinkers are used, animals may need to be trained to use them.

### 4.4. Identification

Ear tags and tattooing should not be used in equines. If identification other than coat colour is required then transponders should be used. Numbered head-collars and hanging tags for halters have also been used successfully for identification.

# H. SPECIES-SPECIFIC GUIDELINES FOR BIRDS

# a. General considerations

# 1. Introduction

Birds are used for a broad range of purposes including fundamental research, applied veterinary medical studies and toxicology. Domestic fowl and turkeys are the most common laboratory birds and are often used in developmental studies and for the production of biological materials such as tissue and antibodies. Domestic poultry are also the most commonly used species in bird welfare research. Fowl are used for pharmaceutical safety and efficacy evaluation, whereas quail and other birds are more frequently the subjects of ecotoxicology studies.

The other, less commonly used species such as the pigeon and wild birds are generally used in psychology and fundamental physiology or zoology research. Catching wild birds to use as experimental animals should be avoided unless it is necessary for the purposes of the experiment.

Although birds are essentially built for flight and share the same basic body plan, they have an extremely diverse range of adaptations for locomotion and feeding. Most species are adapted to range over relatively large, three-dimensional areas by one or more means of locomotion including flying, walking, running, swimming or diving, both while foraging and during migration. Many species of birds are highly social and should be kept in stable groups wherever possible.

Additional details are provided for the commonly bred and used laboratory species. It is essential that the housing and care of less commonly used species not included below pay due regard to their behavioural, physiological and social requirements. Housing, husbandry and care protocols for such species should be researched before birds are obtained or used. Advice on requirements for other species (or if behavioural or breeding problems occur) should be sought from experts and care staff to ensure that any particular species needs are adequately addressed. Information and guidance on less commonly used species is available in the background information document.

During agricultural research when the aim of the research requires that the animals are kept under similar conditions to those under which commercial farm animals are kept, the keeping of the animals should at least follow the standards laid down in Directive 98/58/EC and in the specific Directive for the protection of laying hens (Council Directive 1999/74/EC (5)) as well as in the Recommendations adopted under the Council of Europe Convention for the Protection of Animals kept for Farming Purposes (ETS No 87).

Many of the potential welfare problems specific to birds are associated with inappropriate pecking behaviour. This can be divided into aggressive pecking; feather pecking (where individuals either peck at other birds' feathers or pluck and pull at their own); and pecking at the skin of other birds, which can cause serious suffering and mortality if unchecked. The cause of inappropriate pecking is not always clear, but it is often possible to avoid outbreaks by rearing chicks with access to substrate that enables them to forage and peck appropriately. Chicks of all species should therefore be housed on solid floors with litter.

Prevention is especially important because fowl are attracted to damaged feathers, and the presence of a few feather-pecked birds may therefore lead to the rapid spread of injurious pecking. There are a number of measures that should be employed to avoid outbreaks of injurious pecking wherever possible and to reduce or prevent this behaviour should it occur. These include providing alternative pecking substrates such as foraging substrate, bunches of string, pecking blocks or straw; providing visual barriers; periodically or temporarily lowering the light intensity or using red light; and using light sources that emit UV rays. Anti-pecking sprays are commercially available and can be used to reduce the incidence of injurious pecking in the short term, but it will still be necessary to address the underlying causes of the behaviour. Some strains of domestic bird have been selectively bred so that inappropriate pecking is reduced and such strains should be researched and used wherever possible.

Methods which cause pain or distress, such as very low lighting (i.e. below 20 lux) for prolonged periods, or for physical modifications such as beak trimming, should not be used.

Birds housed in a poor quality environment that does not permit them to forage, exercise or interact with conspecifics will experience chronic distress that may be indicated by stereotypic behaviour, for example self-mutilation, feather pecking, and pacing. Such behaviour may be indicative of serious welfare problems and should lead to an immediate review of housing, husbandry and care.

### The environment and its control

### 2.1. Ventilation

Many species are especially susceptible to draughts. Measures should therefore be in place to ensure that individuals do not become chilled. Accumulation of dust and gases such as carbon dioxide and ammonia should be kept to a minimum.

# 2.2. Temperature

Where appropriate, birds should be provided with a range of temperatures so that they can exercise a degree of choice over their thermal environment. All healthy adult quail, pigeon and domestic ducks, geese, fowl and turkeys should be housed at temperatures between 15 °C and 25 °C. It is essential to take account of the interaction between temperature and relative humidity, as some species will suffer from heat stress within the prescribed temperature range if relative humidity is too high. For species where there are no published guidelines on temperature and humidity, the climate experienced in the wild throughout the year should be researched and replicated as closely as possible. Higher room temperatures than those indicated or a localised source of supplementary heat such as a brooder lamp may be required for sick or juvenile birds (see Table H.1.below).

Table H.1.

Guidelines for temperatures and relative humidities for domestic fowl and turkeys, G. gallus domesticus and Meleagris gallopavo

Age (days)	Under lamp (°C)	Ambient temperature in room (°C)	Relative humidity (%)	
Up to 1	35	25 to 30	60 to 80	
Over 1 to 7	32	22 to 27	60 to 80	
Over 7 to 14	29	19 to 25	40 to 80	
Over 14 to 21	26	18 to 25	40 to 80	
Over 21 to 28	24	18 to 25	40 to 80	
Over 28 to 35	_	18 to 25	40 to 80	
Over 35	_	15 to 25	40 to 80	

The chicks' behaviour should be used as a guide when setting brooder lamp temperature.

If thermally comfortable, chicks of all species will be evenly spaced in the enclosure and making a moderate amount of noise; quiet chicks may be too hot and chicks making noisy distress calls may be too cold.

# 2.3. Humidity

Relative humidity should be maintained within the range of 40 to 80 % for healthy, adult, domestic birds.

# 2.4. Lighting

Light quality and quantity are critically important for some species at certain times of the year for normal physiological functioning. Appropriate light and dark regimes for each species, life stage and time of year should be known before animals are acquired.

Lights should not be abruptly switched off or on, but should be dimmed and raised in a gradual fashion. This is especially important when housing birds capable of flight. Dim night-lights may facilitate movement at night for heavy-bodied poultry strains. Where provided, care should be taken to ensure that circadian rhythms are not disrupted.

# 2.5. Noise

Some birds, for example the pigeon, are considered to be able to hear very low frequency sounds. Although infrasound (sound below 16 Hz) is unlikely to cause distress, birds should be housed away from any equipment that emits low frequency vibrations whenever possible.

# 3. Health

Captive-bred birds should be used wherever possible. Wild birds may present special problems in terms of their behaviour and health when in a laboratory situation. A longer period of quarantine and habituation to captive conditions is generally required before they are used in scientific procedures.

Careful health monitoring and parasite control should minimise health risks in birds with outdoor access.

### 4. Housing, enrichment and care

Birds should be housed in enclosures which facilitate and encourage a range of desirable natural behaviours, including social behaviour, exercise and foraging. Many birds will benefit from housing that allows them to go outdoors and the feasibility of this should be evaluated with respect to the potential to cause distress or to conflict with experimental aims. Some form of cover such as shrubs should always be provided outdoors to encourage birds to use all the available area.

# 4.1. Housing

Birds should be housed in socially harmonious groups within the animal enclosure, unless the scientific procedures or welfare requirements make this impossible. Special care is needed when regrouping birds or introducing an unfamiliar bird to a group. In all cases, groups should be monitored for social compatibility on an ongoing basis.

Single-housing of birds for even short periods can be a significant stress factor. Therefore, birds should not be single-housed unless justified on welfare or veterinary grounds. Single-housing on experimental grounds should be determined in consultation with the animal technician and with the competent person charged with advisory duties in relation to the well-being of the animals.

Most species of bird are social for at least part of the year and highly sensitive to family relationships, so the formation of appropriate, stable, harmonious groups should be given a high priority. As there are significant species variations, the optimal composition of groups, and at what stage in the birds' lives these should be created should be known before groups are formed and procedures are undertaken.

### 4.2. Enrichment

A stimulating environment is a very important contributor to good bird welfare. Perches, dust and water baths, suitable nest sites and nesting material, pecking objects and substrate for foraging should be provided for species and individuals that will benefit from them unless there is scientific or veterinary justification for withholding such items. Birds should be encouraged to use all three dimensions of their housing for foraging, exercise and social interactions including play wherever possible.

# 4.3. Enclosures — dimensions and flooring

Guidelines for enclosure dimensions are set out in the species-specific guidelines for domestic fowl, domestic turkeys, quail, ducks and geese, pigeons and zebra finches. All birds, especially species that spend a significant proportion of their time walking, such as quail or fowl, should be housed on solid floors with substrate rather than on grid floors. Birds can be prone to foot problems, for example, overgrown claws, faecal accumulation and foot lesions such as foot-pad dermatitis due to standing on wet litter, on any type of flooring, and so frequent monitoring of foot condition is always necessary. In practice, it may be necessary to consider a compromise between solid-and grid flooring for scientific purposes. In such cases, birds should be provided with solid-floored resting areas occupying at least a third of the enclosure floor. Grid areas should be located under perches if faecal collection is required. To reduce the incidence of foot injuries, slats made of plastic should be used in preference to wire mesh wherever possible. If wire mesh has to be used, it should be of a suitable grid size to adequately support the foot and the wire should have rounded edges and be plastic coated.

# 4.4. Feeding

Feeding patterns of wild birds vary widely and consideration should be given to the nature of the food, the way in which it is presented and the times at which it is made available. Diets that will meet the nutritional requirements of each species and promote natural foraging behaviour should be researched and formulated before any animals are obtained. Part of the diet or additional treats should be scattered on the enclosure floor to encourage foraging wherever appropriate. Dietary enrichment benefits birds, so additions such as fruit, vegetables, seeds or invertebrates should be considered where appropriate even if it is not possible to feed birds on their 'natural' diet. Where new foods are introduced, the previous diet should always be available so that birds will not go hungry if they are unwilling to eat new foods. Some species are more adaptable than others and advice should be sought on appropriate dietary regimes.

As some species, particularly granivores, require grit to digest their food, these should be provided with appropriately-sized grit. Birds will select grit of the size they prefer if material of various sizes is provided. The grit should be renewed regularly. Dietary calcium and phosphorus should also be provided for birds in an appropriate form and at an appropriate level for each life stage, to prevent nutritional bone disease. Any such requirements should be thoroughly researched and catered for. Food can be supplied in feeders that are either attached to the side of the enclosure or standing on the enclosure floor. Space occupied by floor feeders is not available to the birds and should not be included in calculations of pen area. Wall mounted feeders do not occupy floor space but should be designed and fitted with care so that birds cannot become trapped underneath them. Chicks of some species (for example, domestic turkeys) may need to be taught to feed and drink in order to avoid dehydration and potential starvation. Food for all species should be clearly visible and provided at several points to help prevent feeding problems.

# 4.5. Watering

Water should be provided via nipple or cup drinkers, or as a continuous drinking channel. There should be sufficient drinkers or an adequate length of channel drinker to prevent dominant birds from monopolising them. One nipple or cup drinker should be provided for every three or four birds, with a minimum of two in each enclosure. Supplementary water may also be given as enrichment in birds' feed if appropriate.

### 4.6. Substrate, litter, bedding and nesting material

Suitable substrates for birds should be absorbent, unlikely to cause foot lesions and of an appropriate particle size to minimise dust and prevent excessive accumulation on the birds' feet. Suitable substrates include chipped bark, white wood shavings, chopped straw or washed sand, but not sandpaper. Litter should be maintained in a dry, friable condition and be sufficiently deep to dilute and absorb faeces. Other suitable floor coverings include plastic artificial turf or deep pile rubber mats. A suitable pecking substrate such as pieces of straw should be scattered over the floor.

Hatchlings and juvenile birds should be provided with a substrate that they can grip to avoid developmental problems such as splayed legs. Juvenile birds should also be encouraged if necessary, for instance by tapping with the fingers, to peck at the substrate to help prevent subsequent misdirected pecking.

# 4.7. Cleaning

(See paragraph 4.9. of the General section).

# 4.8. Handling

Suitable equipment for catching and handling should be available, for example, well maintained nets in appropriate sizes and darkened nets with padded rims for small birds.

If the experimental procedure requires adult birds to be handled regularly, it is recommended from a welfare and experimental perspective to handle chicks frequently during rearing, as this reduces later fear of humans.

# 4.9. Humane killing

The preferred method of killing for juvenile and adult birds is an overdose of anaesthetic using an appropriate agent and route. This is preferable to carbon dioxide inhalation, as carbon dioxide may be aversive.

As diving birds and some others, for example, mallard ducks, can slow their heart rates and hold their breath for long periods, care should be taken when killing such species by inhalation to ensure that they do not recover. Ducks, diving birds and very young chicks should not be killed using carbon dioxide.

### 4.10. Records

(See paragraph 4.12. of the General section)

# 4.11. Identification

Non-invasive or minimally invasive methods such as noting physical differences, ringing with either closed or split rings and staining or dyeing the feathers are preferable to more invasive techniques such as electronic tagging or wing tagging. Combinations of coloured leg rings minimise handling for identification, although due regard

should be paid to any potential impact of colours on behaviour in some species. When using rings as temporary marking for rapidly growing chicks, regular checking is essential to ensure that the ring is not impeding the growth of the leg.

Highly invasive marking methods such as toe-clipping or web-punching cause suffering and should not be used.

### b. Additional guidelines for housing and care of the domestic fowl, in stock and during procedures

Domestic fowl (*Gallus gallus domesticus*) retain much of the biology and behaviour of the Jungle fowl from which they were domesticated. Behaviours that are most important to the species are nesting (in females), perching and using litter for foraging, scratching, pecking and dustbathing. Fowl are social and should be housed in groups of around five to twenty birds, with fewer males than females in adult groups, for example, a ratio of 1 to 5. Attempts have been made to select strains of fowl for reduced feather pecking or agonistic behaviour. The existence of appropriate strains of this type should be determined, and the feasibility of acquiring them, should be assessed for each project.

Laying hens should have access to nest boxes from at least two weeks before coming into lay and no later than 16 weeks of age. Single- or pair-housed birds should each have access to a nest box, with a ratio of at least one nest box per two birds provided in larger groups. Nest boxes should be enclosed and large enough to allow one hen to turn around. A loose substrate such as wood-shavings or straw should be supplied within nest boxes to promote nesting behaviour. Substrate should be regularly replaced and kept clean.

Fowl should always be provided with the opportunity to perch, peck appropriate substrates, forage and dust-bathe from one day old. Suitable materials for dust-bathing include sand or soft wood shavings.

Perches should be 3 to 4 cm in diameter and round with a flattened top. The optimum height above the floor varies for different breeds, ages and housing conditions but perches should initially be fixed at 5 to 10 cm and for older birds at 30 cm above the floor. Perch heights should be adjusted in response to the birds' behaviour by seeing how easily birds can get on and off perches and move between them. All birds should be able to perch at the same time and every adult bird should be allowed 15 cm of perch at each level. Especially during the establishment of groups, birds should also be briefly observed during dark periods to confirm that all individuals are roosting.

Fowl are highly motivated to perform 'comfort behaviour' such as wing flapping, feather ruffling and leg stretching, which help to maintain strong leg bones. Birds should therefore be housed in floor enclosures large enough to permit all of these behaviours whenever possible. Ideally, birds should be housed with outdoor access; appropriate cover such as bushes is essential to encourage fowl to go outside.

Flooring for fowl should be solid, as this enables the provision of substrate to encourage foraging and possibly help to reduce the incidence of feather pecking. If fowl need to be caged for scientific purposes, they should be housed in enclosures designed to address behavioural requirements. If there are scientific reasons for not providing a solid floor, a solid area with loose substrate and items such as bunches of string, pecking blocks, rope, turf or straw should be provided for pecking.

Fowl strains developed for rapid growth rates (broilers) are highly susceptible to lameness and their use should be avoided wherever possible. If broilers are used, individuals should be assessed for lameness at least weekly and grown more slowly than those reared commercially unless growth rate is essential for the study.

Table H.2.

Domestic fowl: Minimum enclosure dimensions and space allowances

Body mass (g)	Minimum enclosure size (m²)	Minimum area per bird (m²)	Minimum height (cm)	Minimum length of feed trough per bird (cm)
Up to 200	1,00	0,025	30	3
over 200 to 300	1,00	0,03	30	3
over 300 to 600	1,00	0,05	40	7
over 600 to 1 200	2,00	0,09	50	15
over 1 200 to 1 800	2,00	0,11	75	15
over 1 800 to 2 400	2,00	0,13	75	15
over 2 400	2,00	0,21	75	15

Where these minimum enclosures sizes cannot be provided for scientific reasons, the duration of the confinement should be justified by the experimenter and determined in consultation with the animal technician and with the competent person charged with advisory duties in relation to the well-being of the animals. In such circumstances, birds can be housed in smaller enclosures containing appropriate enrichment and with a minimum floor area of  $0.75 \, \text{m}^2$ . These can be used to house two laying birds or small groups of birds in accordance with the space allowances given above.

# c. Additional guidelines for housing and care of the domestic turkey, in stock and during procedures

Wild turkeys regularly utilise a diverse range of environments and perform a variety of behaviours including dust-bathing, foraging and hunting. The social behaviour of the wild turkey is complex, particularly during the breeding season. Domestic turkeys (*Meleagris gallopavo*) retain many of the characteristics of wild birds but there are some fundamental differences, for example domestic turkeys are unable to fly but have retained the ability to run quickly, and jump and glide, especially at younger ages.

Domestic turkeys are highly social and should not be single-housed. Stable groups should be formed as soon as birds are acquired and adequate monitoring is essential as injurious feather-pecking and head-pecking can occur from the first day of life.

Lameness is a common problem and needs to be carefully monitored. Veterinary advice should be sought on a policy for dealing with lameness.

Turkeys should be provided with perches placed at a height where birds on the ground are not able easily to peck and tug at the feathers of perching birds. However, if birds are older and less agile, the access to perches should be facilitated by special equipment such as ramps. Where this is not possible, perches should be placed at a low height (for example at 5 cm). The shape and size of the perch should be in accordance with the rapidly growing claws of the birds. Perches should be ovoid or rectangular with smoothed corners and made of wood or plastic.

Substrate for dust-bathing should always be provided. Suitable materials are fresh sawdust or sand. Straw bales may be used for enrichment and to provide a refuge from dominant birds, but will need to be frequently replaced and older, heavier birds may need ramps to gain access to them.

Table H.3.

Domestic Turkey: Minimum enclosure dimensions and space allowances

Body mass (kg)	Minimum enclosure size (m²)	Minimum area per bird (m²)	Minimum height (cm)	Minimum length of feed trough per bird (cm)
Up to 0,3	2,00	0,13	50	3
over 0,3 to 0,6	2,00	0,17	50	7
over 0,6 to 1	2,00	0,30	100	15
over 1 to 4	2,00	0,35	100	15
over 4 to 8	2,00	0,40	100	15
over 8 to 12	2,00	0,50	150	20
over 12 to 16	2,00	0,55	150	20
over 16 to 20	2,00	0,60	150	20
over 20	3,00	1,00	150	20

All enclosure sides should be at least 1,5 m long. Where these minimum dimensions cannot be provided for scientific reasons, the duration of the confinement should be justified by the experimenter and determined in consultation with the animal technician and with the competent person charged with advisory duties in relation to the well-being of the animals. In such circumstances, birds can be housed in smaller enclosures containing appropriate enrichment and with a minimum floor area of  $0.75 \, \text{m}^2$  and a minimum height of 50 cm for birds below  $0.6 \, \text{kg}$ ,  $75 \, \text{cm}$  for birds below  $4 \, \text{kg}$ , and  $100 \, \text{cm}$  for birds over  $4 \, \text{kg}$ . These can be used to house small groups of birds in accordance with the space allowances given above.

# d. Additional guidelines for housing and care of quail, in stock and during procedures

Wild quail live in small social groups and devote much of their time to scratching and foraging for seeds and invertebrates on the ground. The preferred habitat of many species is dense vegetation such as grasslands, bushes alongside rivers and cereal fields. Domestication does not appear substantially to have altered quail behaviour, so it is essential to design housing systems that respect this and allow the provision of substrate for scratching, pecking and dustbathing, nestboxes and cover wherever possible. The housing of quail in aviaries or pens as opposed to cages is therefore strongly recommended.

Quail (Coturnix spp; Colinus virginianis; Lophortyx californica; Excalfactoria chinensis) should be group housed in either all female or mixed-sex groups. Where the sexes are mixed, the ratio of males to females should be low (for example, 1 to 4) to reduce aggression between males and injuries to females. It may be possible to pair-house males if stable pairs are formed during rearing. The likelihood of aggressive pecking leading to skin lesions and feather loss is reduced if quail are not kept under intensive conditions and established groups are not mixed.

Quail are capable of extremely rapid startle responses, which can lead to head injuries. Staff should therefore always approach birds slowly and calmly and quail should be provided with cover and environmental enrichment, especially early in life, in order to reduce fear. Quail chicks should have access to coloured objects such as balls, tubing and cubes to alleviate fear of both human beings and novel stimuli in adult birds. Adult birds should be given pecking objects such as stones, pine cones, balls and branches of vegetation. Sand, wood shaving or straw substrate for foraging and a place to which the birds can withdraw should be provided, with additional dust baths of sand or sawdust if the foraging substrate is not suitable for dust bathing. Laying hens should have access to nest boxes and nesting material, such as hay.

If quail need to be housed in cages, consideration should be given to combining enclosures and adding enrichment items. Solid enclosure roofs may make birds feel safer, although this could result in unacceptably low light levels in lower enclosures if birds are housed in racks. Birds should be cage-housed for the minimum possible period because many welfare problems become more severe with age, especially in birds kept for one year or more.

Table H.4.

Quail: Minimum enclosure dimensions and space allowances

Body mass (g)	Minimum enclosure size (m²)	Area per bird pair-housed (m²)	Area per additional bird group-housed (m²)	Minimum height (cm) (*)	Minimum length of trough per bird (cm)
Up to 150	1,00	0,5	0,10	20	4
over 150	1,00	0,6	0,15	30	4

<sup>(\*)</sup> The enclosure roof should be made of pliant material to reduce the risk of head injuries.

#### e. Additional guidelines for housing and care of ducks and geese, in stock and during procedures

Domestic ducks and geese commonly used in research and testing include Anas platyrhynchos, Anser anser domesticus and Cairina moschata. All waterfowl are primarily adapted for locomotion and feeding in water, which is also very important for 'comfort' behaviours such as bathing and preening. Ducks and geese should be provided with a pond with a mixture of stones and grit on the bottom, both to increase the birds' behavioural repertoire and to encourage adequate maintenance of the feathers. The very minimum that waterfowl should be able to do is immerse their heads under water and shake water over the body. Drinkers and ponds for waterfowl should be located over grid areas with drains beneath to reduce flooding.

Domestic geese and ducks have been selected for meat and egg production, but all breeds retain most of their 'wild type' behaviour and are generally more nervous and easily upset than other domestic birds, especially when they are moulting.

Within twenty-four hours of hatching and throughout the first week of life, water should be provided to facilitate swimming behaviour, but care should be taken to minimise the risk of drowning by, for example, the use of a shallow bowl. After the first week, a shallow pond (dimensions as in table H.5) with large stones on the bottom should be provided with food or grit scattered among the stones to encourage dabbling or diving, as appropriate. In the absence of the parent birds, access to ponds for juvenile birds should only be under supervision to ensure that they can leave the water and do not become chilled. This should continue until they are clearly capable of leaving the water unaided and their waterproof feathers have begun to emerge. It is not necessary to control the temperature of the water. Ponds should be regularly cleaned and water replaced as necessary to ensure good water quality.

Ducks and geese should be housed on solid floors and have sufficient space to permit foraging, walking, running and wing flapping. A complex environment should be provided, including for example natural or artificial cover, boxes and straw bales. Ducks and geese should always be kept outdoors or have access to outdoor runs unless there is scientific or veterinary justification for keeping them indoors. Birds housed with outside access should be kept secure from predators and should be supplied with a dry shelter to enable them to rest. Vegetation for cover and/or grazing should be provided as applicable. Serious consideration should be given to supplying other features of the habitat that are likely to be important to each species whether birds are housed indoors or outdoors. This includes shallow water with vegetation for dabbling ducks, turf for geese and deeper water with large stones for species whose natural habitat is along rocky coastlines.

Ducks and geese should be housed in appropriately sized groups wherever possible and the amount of time when any individual is left alone should be minimised. Many species become territorial during the breeding season, however, so it may be necessary to reduce group sizes and ensure that there is sufficient enclosure space to reduce the risk of injury, particularly to female birds.

Table H.5. Ducks and geese: Minimum enclosure dimensions and space allowances

Body mass (g)	Minimum enclosure size (m²)	Area per bird (m²) (*)	Minimum height (cm)	Minimum length of feed trough per bird (cm)
Ducks				
Up to 300	2,00	0,10	50	10
over 300 to 1 200 (**)	2,00	0,20	200	10
over 1 200 to 3 500	2,00	0,25	200	15
over 3 500	2,00	0,50	200	15
Geese				
Up to 500	2,00	0,20	200	10
over 500 to 2 000	2,00	0,33	200	15
over 2 000	2,00	0,50	200	15

 <sup>(\*)</sup> This should include a pond of minimum area 0,5 m² per 2 m² enclosure with a minimum depth of 30 cm. The pond may contribute up to 50 % of the minimum enclosure size.
 (\*\*) Pre-fledged birds may be held in enclosures with a minimum height of 75 cm.

Where these minimum enclosure sizes cannot be provided for scientific reasons, the duration of the confinement should be justified by the experimenter and determined in consultation with the animal technician and with the competent person charged with advisory duties in relation to the well-being of the animals. In such circumstances, birds can be housed in smaller enclosures containing appropriate enrichment and with a minimum floor area of 0,75 m<sup>2</sup>. These can be used to house small groups of birds in accordance with the space allowances given below.

#### f. Additional guidelines for housing and care of pigeons, in stock and during procedures

The various strains of domestic pigeon are believed to derive from the rock dove Columbia livia. Rock doves nest and roost on cliffs or within caves, and feral pigeons will utilise sheltered ledges on man-made structures in the same way. In their natural habitat pigeons usually occur in pairs to large flocks, feeding and roosting together, but will defend roosting spaces and nesting areas. Pigeons can be housed in mixed groups, and may lay eggs but will not incubate them if nest boxes are not provided.

Care should be taken when choosing a breed for laboratory use, as some strains may show abnormal or undesirable behaviours and should therefore be avoided. Pigeons are primarily seed-eaters but are omnivorous, so food containing animal protein should be offered regularly.

Pigeons should be allowed an area sufficient for flight wherever possible, with a separate perching area for each bird along at least one wall of the enclosure. Box perches approximately 30 cm × 15 cm located in blocks should be provided. Branches hung from the roof and scaffolding can also be used for perching. Toys hung from chains should be provided, for example, bird bells, mirrors and commercially available toys designed for pets. Each enclosure should have shallow water baths. Where pigeons need to be handled frequently, 'nesting areas' or chambers can be provided so that birds can be trained to retreat to them for capture.

Larger, enriched enclosures with shelving, perches and toys should be used wherever possible rather than 'standard' pigeon enclosures. Pigeons benefit from being able to forage and should not be kept on grid floors without strong scientific justification.

Table H.6.

Pigeons: Minimum enclosure dimensions and space allowances

Group size	Minimum enclosure size (m²)	Minimum height (cm)	Minimum length of food trough per bird (cm)	Minimum length of perch per bird (cm)
Up to 6	2	200	5	30
from 7 to 12	3	200	5	30
For each additional bird above 12	0,15		5	30

Enclosures should be long and narrow (for example 2 m by 1 m) rather than square to allow birds to perform short flights.

# g. Additional guidelines for housing and care of zebra finch, in stock and during procedures

Zebra finches (*Taeniopygia guttata*) occur across most of Australia. They are highly mobile, ranging over wide areas in search of food, and live in flocks of up to several hundred individuals. The species is monogamous and sexually dimorphic, as the male's plumage is more ornate than that of the female. The breeding season is not fixed, but is triggered by the availability of ripening grass seeds. Zebra finches use nests for roosting as well as breeding; roosting nests are used more frequently in cold conditions and may be old breeding nests or purpose-built.

Zebra finches are social and non-breeding birds should be housed in groups. Unwanted breeding can be prevented by housing in single sex groups, or suppressed in mixed-sex groups by withholding both roosting and breeding nests and by feeding a diet of dry seeds supplemented with fresh greens, but never soaked or sprouted seeds. Nests should be provided for breeding birds, for example in the form of wicker or plastic baskets or wooden boxes with dried grass, paper strips or coconut fibres for nesting material, but birds will defend these and it is important to monitor behaviour to ensure that sufficient nests are provided. Sprays of *Panicum* millet should be continually available as dietary enrichment. As zebra finches feed extensively on the ground, birds should be housed on solid floors to facilitate natural foraging behaviour.

Toys, perches and swings designed for pet birds will benefit zebra finches and these should be provided wherever possible. Perches are particularly important for well-being and should be provided at a range of heights to facilitate normal feeding and roosting behaviour. Water for bathing should be provided at least once a week in shallow trays with water of approximately 0,5 to 1 cm in depth.

Fitting zebra finches with coloured leg bands for identification can have significant effects on their social and reproductive behaviour (for example, red can enhance dominance and green or blue reduce it). Care should be taken in the selection of colours and patterns of leg bands.

Minimum enclosure sizes for zebra finches are set out in Table H.7 below. Enclosures should be long and narrow (for example, 2 m by 1 m) to enable birds to perform short flights. Zebra finches thrive in outdoor enclosures provided they have access to shelter and roosting nests where appropriate. Additional heating should be provided for birds housed outdoors in cold conditions.

Table H.7.

Zebra Finch: Minimum enclosure dimensions and space allowances

Group size	Minimum enclosure size (m²)	Minimum height (cm)	Minimum number of feeders	
Up to 6	1,0	100	2	
7 to 12	1,5	200	2	
13 to 20	2,0	200	3	
for each additional bird above 20	0,05		1 per 6 birds	

For breeding studies, pairs may be housed in smaller enclosures containing appropriate enrichment with a minimum floor area of  $0.5 \text{ m}^2$  and a minimum height of 40 cm. The duration of the confinement should be justified by the experimenter and determined in consultation with the animal technician and with the competent person charged with advisory duties in relation to the well-being of the animals.

#### I. SPECIES-SPECIFIC GUIDELINES FOR AMPHIBIANS

#### 1. **Introduction**

According to systematics, amphibians involve three main orders: *Urodela (Caudata), Gymnophiona (Apoda)*, and *Anura (Ecaudata)*. The *Anura* belong to the super-order *Salientia*. For the present guidelines, *Urodela* (salamanders, newts) and *Anura* (frogs, toads) are of interest. They differ greatly in their patterns of geographic distribution and in the diversity of living types, such as aquatic (for example, *Xenopus laevis*), semi-aquatic (for example, *Rana temporaria*), semi-terrestrial (for example, *Bufo marinus*) and arboreal (for example, *Hyla cinerea*). Amphibians occupy a wide range of habitat types from arid deserts to deep freshwater lakes. Some may spend most of their life underground or high in cloud forest canopy. Some are found north of the Arctic Circle and can tolerate freezing conditions, while others have evolved a range of adaptations to avoid desiccation in hot areas of the world.

Amphibians are very much adapted to the substrate on/in which they live. In this context, the body skin plays an important role in the transfer of water, soluble substances, including toxic substances and oxygen. Therefore it plays key roles in the survival of amphibians, their interaction with their environment, and their ability to exploit a wide range of habitats and ecological conditions. An amphibian's health depends on certain properties and peculiarities of its body skin, thus making amphibians significant bio-indicators of environmental health.

Where possible, amphibians used for experimental or other scientific purposes should be bred and reared in captivity. Purpose-bred animals should be used in preference to animals taken from the wild.

Table I.1. lists the four main habitats of amphibians and examples of species of each habitat frequently used for experimental and other scientific purposes. The following proposals provide details on the basic accommodation and care conditions to be covered for species of these habitats. Specific procedures may require the use of certain other species which do not fall into the four habitat categories. Further advice on requirements for these and other species (or if behavioural or breeding problems occur) should be sought from expert specialists and care staff to ensure that any particular species needs are adequately addressed. Additional background information on less commonly used species, and habitats is available in the background information document elaborated by the Group of Experts.

Table I.1:

Main habitat categories and examples by habitat of species frequently used

Habitat	Amphibian species	Size (cm)	Original geographic distribution/Biotope	Optimal temperature	Relative humidity	Main period of activity
Aquatic (Urodeles)	Ambystoma mexi- canum (Axolotl)	24 to 27	Mexico/Channels of the former sea of Xochimilco	15 °C to 22 °C	100 %	Twilight
Aquatic (Anurans)	Xenopus laevis (Clawed frog)	6 to 12	Central and South Africa/ponds, ground water and spring-fed	18 °C to 22 °C	100 %	Twilight/ night
Semi-aquatic (Anurans)	Rana temporaria (Common frog)	7 to 11	Europe (middle and north) to Asia (without southern Balkan)/Near ponds, lakes, streams (shores, meadows)	10 °C to 15 °C	50 to 80 %	Day/night

Habitat	Amphibian species	Size (cm)	Original geographic distribution/Biotope	Optimal temperature	Relative humidity	Main period of activity
Semi-terrestrial (Anurans)	Bufo marinus (Marine toad)	12 to 22	Central and South America/Man- grove, woods	23 °C to 27 °C	50 to 80 %	Night
Arboreal (Anurans)	Hyla cinerea (Green tree frog)	3 to 6	Southeast USA/ Open bushy bor- ders of cypress swamps, flat coun- try, forest	18 °C to 25 °C	50 to 70 %	Day/night

# 2. The environment and its control

### 2.1. Ventilation

Enclosures for amphibians should be adequately ventilated. The water in enclosures of aquatic caged amphibians should be filtered, circulated, and aerated (see also paragraph 4.3.1.).

## 2.2. Temperature

Amphibians are ectothermic. Areas of different temperature and humidity are beneficial, to allow amphibians to seek their preferred microenvironment. Amphibians exposed to frequent fluctuations in temperature and humidity may be severely stressed and may be more prone to health problems. Room and water temperatures should be controlled.

Hibernation in amphibians may be induced or interrupted by regulating light-dark rhythm and room temperature. Before inducing hibernation in captivity, animals should be in good health and body condition. In animals used for breeding, a state of near winter torpor (for example dim light to darkness and 8  $^{\circ}$ C to 10  $^{\circ}$ C room temperature) may be simulated where appropriate. Under these conditions, the animals can be kept without feeding for as long as four to five months. Restoration of pre-hibernation environmental conditions will induce activity and mating behaviour.

Prevention of hibernation in a laboratory environment will not cause major welfare problems.

### 2.3. Humidity

Amphibians do not drink but absorb moisture through their skin. Water loss is an especially critical problem in captive terrestrial and semi-terrestrial amphibians, as a properly hydrated integument is essential to the normal function of the amphibian skin. Areas of different humidity within the enclosure are beneficial. Even desert-adapted amphibians should have access to a humid environment.

# 2.4. Lighting

Photoperiods reflecting the natural cycle from where the animals originate should be used. Light levels in the enclosures should be consistent with that expected to be encountered under natural conditions. Both semi-terrestrial and aquatic caged animals should have the opportunity to withdraw to shaded areas within the enclosure.

## 2.5. Noise

Amphibians are very sensitive to noise (airborne stimuli) and vibration (substrate-borne stimuli) and are disturbed by any new, unexpected stimulus. Therefore, such extraneous disturbances should be minimised.

### 2.6. Alarm systems

Adequate alarm systems are recommended if circulation systems are used and/or aeration is required.

#### 3. Health

(See paragraph 4.1. of the General section)

## 4. Housing, enrichment and care

#### 4.1. Housing

In most amphibians, social behaviour is mainly restricted to the mating season. However, group-housing of amphibians is advisable, for instance to improve feeding and reduce fear responses. For example, in Xenopus spp. group feeding promotes feeding frenzies inducing all animals to feed. At very low stocking densities such frenzies do not occur and food is frequently not eaten.

To avoid cannibalism in certain species (particularly among larval *Ambystoma* spp. and *Scaphiopus* spp.), these animals should be maintained in small groups. Cannibalism in groups can be reduced by size grading.

#### 4.2. Enrichment

The terrestrial habitat of amphibians should be structured, including, for example, branches, leaves, pieces of bark, stones or other suitable man-made materials. Amphibians benefit from such environmental enrichment in different ways: for example, such inclusions allow animals to hide, and provide labels for visual and spatial orientation. The side walls of the terraria should be textured to provide a structured surface.

The provision of hiding places/shelters that are appropriate to the amphibian's needs is recommended, because they can reduce stress on captive amphibians. For example, in *Xenopus* spp. a tube of ceramic or plastic may be provided. Refuges should be inspected regularly for sick or injured animals. A dark floor to the tank may enhance the sense of security in the animals.

Materials used for enrichment devices should not be detrimental to the health of the amphibians. Enclosures and enrichment structures should have smooth surfaces and rounded edges to minimise the risk of injury to the amphibian's skin.

# 4.3. Enclosures — dimensions and flooring

## 4.3.1. Enclosures for aquatic amphibians

Aquatic amphibians such as *Xenopus laevis* or amphibian larvae are housed in tanks and aquaria. These may be equipped with a gentle flow-through water system for the circulation of uncontaminated (for example, dechlorinated) water, a heating device to maintain suitable temperatures, and a compressed air supply and airstones for aeration. Care is needed to ensure that aeration does not cause injury to the animals. Unless a proper flow system is in place, the water in the enclosures should be renewed with water of an appropriate quality about twice a week.

For *Xenopus* spp., systems with regular changes of water (fill-and-dump systems) are sufficient for maintaining appropriate water quality (such as minimising levels in ammonia). Airstones are not required for *Xenopus*.

Furthermore, long, narrow enclosures should be avoided since they may restrict locomotor activity and social behaviour such as feeding frenzies.

Table 1.2.

Aquatic urodeles, e.g., Ambystoma spp: Minimum enclosure dimensions and space allowances

Body length (*) (cm)	Minimum water surface area (cm²)	Minimum water surface area for each additional animal in group-holding (cm²)	Minimum water depth (cm)
Up to 10	262,5	50	13
over 10 to 15	525	110	13
over 15 to 20	875	200	15

Body length (*) (cm)	Minimum water surface area (cm²)	Minimum water surface area for each additional animal in group-holding (cm²)	Minimum water depth (cm)
over 20 to 30	1 837,5	440	15
over 30	3 150	800	20
(*) measured from snout to tail			

Table I.3. Aquatic anurans, e.g., Xenopus spp: Minimum enclosure dimensions and space allowances. (\*\*)

Body length (**) (cm)	Minimum water surface area (cm²)	Minimum water surface area for each additional animal in group-holding (cm²)	Minimum water depth (cm)
Less than 6	160	40	6
from 6 to 9	300	75	8
over 9 to 12	600	150	10
over 12	920	230	12,5

these recommendations apply to holding (i.e., husbandry) tanks but not to those tanks used for natural mating and super-ovulation for reasons of efficiency, as the latter procedures require smaller individual tanks. Space requirements determined for adults in the indicated size categories; juveniles and tadpoles should either be excluded, or dimensions altered according to the scaling principle
(\*\*) measured from snout to vent

#### 4.3.2. Enclosures for semi-aquatic and semi-terrestrial amphibians

Semi-aquatic and semi-terrestrial amphibians are kept in enclosures consisting of a terrestrial part and a aquatic part. The water area of the terrarium should allow animals to submerge. Unless a flow-through system is used, water should be renewed at least twice a week.

Each terrarium should be covered to prevent escape. It is advisable to paint or otherwise cover the outside of transparent walls to minimise damage to the animal. Additions to the interior design can include: soft-foamed plastic material on the floor near the pool area, stones, pieces of artificial bark material, artificial branches and leaves, and shelves. Fine sawdust and any other related small-particle substrate should be avoided, as it affects the sensitive body skin, harbours pathogens and is difficult to clean and re-use.

Table I.4. Semi-aquatic anurans, e.g., Rana temporaria: Minimum enclosure dimensions and space allowances

Body length (*) (cm)	Minimum enclosure size (**) (cm²)	Minimum area for each additional animal in group holding (cm²)	Minimum enclosure height (***) (cm)	Minimum water depth (cm)
Up to 5,0	1 500	200	20	10
over 5,0 to 7,5	3 500	500	30	10
over 7,5	4 000	700	30	15

measured from snout to vent

one third land division, two thirds water division sufficient for animals to submerge measured from the surface of the land division up to the inner part of the top of the terrarium; furthermore, the height of the enclosures should be adapted to the interior design

Table I.5. Semi-terrestrial anurans, e.g., Bufo marinus: Minimum enclosure dimensions and space allowances

Body length (*) (cm)	Minimum enclosure size (**) (cm²)	Minimum area for each additional animal in group- holding (cm²)	Minimum enclosure height (***) (cm)	Minimum water depth (cm)
Up to 5,0	1 500	200	20	10
over 5,0 to 7,5	3 500	500	30	10
over 7,5	4 000	700	30	15

#### 4.3.3. Enclosures for arboreal amphibians

Having regard for the behaviour of different arboreal species, every effort should be made to allow for this by the provision of appropriate structures for climbing and resting by arboreal species (see section 4.3.2). In addition, it is necessary to provide water in which they can submerge themselves or seek greater humidity. If water dishes are used, they should be arranged in such a way that they are easy for the amphibians to enter or to leave.

Table I.6: Arboreal anurans, e.g., Hyla cinerea: Minimum enclosure dimensions and space allowances

Body length (*) (cm)	Minimum enclosure size (**) (cm²)	Minimum area for each additional animal in group-holding (cm²)	Minimum enclosure height (***) (cm)
Up to 3,0	900	100	30
over 3,0	1 500	200	30

measured from snout to vent

#### 4.4. Feeding

The majority of amphibians are carnivores with food preferences for living small invertebrates (such as larvae, insects and worms). Captive animals should be maintained on their natural foods or on foodstuffs approximating those of their natural diets. However, captive aquatic amphibians can successfully be maintained on pieces of fish fillet or scrapings from frozen liver and heart. The feeding frequency should be related to environmental conditions, such as temperature and light intensity. Daily feeding is not advisable for adult animals, but once to three times weekly to satiation at each feeding is recommended.

#### 4.5. Water quality

For aquatic and semi-aquatic amphibians water quality, including the concentration of ammonia and the pH level in water, should be regularly monitored.

#### 4.6. Substrate, litter, bedding and nesting material

(See paragraph 4.8 of the General section)

measured from snout to vent two-thirds land division, one-third water division sufficient for animals to submerge measured from the surface of the land division up to the inner part of the top of the terrarium; furthermore, the height of the enclosures should be adapted to the interior design

two-thirds land division, one-third pool division sufficient for animals to submerge measured from the surface of the land division up to the inner part of the top of the terrarium; furthermore, the height of the enclosures should be adapted to the interior design including, e.g., shelves, large artificial branches, and structures for

## 4.7. Cleaning

In order to avoid diseases, the terrestrial and aquatic areas in the terraria should be carefully cleaned to remove dirt, excrement and food particles.

## 4.8. Handling

The skin of amphibians can be easily damaged. Care is required during handling, which should be kept to a minimum.

#### 4.9. Anaesthesia and humane killing

Invasive, potentially painful procedures should be accompanied both by analgesia and anaesthesia. As amphibians' skin accounts for a significant portion of normal gaseous exchanges, in anaesthetised animals, in which lung respiration is reduced or interrupted, the body skin should always be kept moist, for example with a wet tissue.

#### 4.10. Records

(See paragraph 4.12. of the General section)

## 4.11. Identification

Where animals need to be identified individually there are a number of suitable methods such as transponders; tank labels for individually housed animals; monitoring pigment or wart configurations; small labels by coloured thread. Chemical markings should not be used, since substances are absorbed through the skin, possibly causing toxic effects. Toe clipping is deleterious and should not be carried out.

## Transport

During transport, amphibians should be provided with sufficient air and moisture and, if necessary, appropriate devices to maintain the required temperature and humidity.

## J. SPECIES-SPECIFIC GUIDELINES FOR REPTILES

## 1. **Introduction**

According to morphological systematics, reptiles include the main orders *Rhynchocephalia* (tuatara), *Squamata* (lizards, snakes), *Chelonia* (tortoises, turtles, and terrapins), and *Crocodilia* (alligators, crocodiles, caimans, and gavials). They differ greatly in their patterns of geographic distribution and in the diversity of living types.

In contrast to the more or less smooth and moist skin seen in amphibians, reptiles have a skin protected by overlapping scales (snakes, lizards), by a box-like shell (chelonians), or by bone plates in the skin (crocodiles, alligators, and caimans). The thick skin is an adaptation to better protect reptiles from the water loss that occurs with the permeable skin of amphibians.

Table J.1. lists two very general habitat categories of reptiles and examples of species of each habitat frequently used for experimental and other scientific purposes. The following proposals provide details on the basic housing and care conditions recommended for species found within these habitats. Specific procedures may require the use of certain other species which do not fall into these categories, such as semi-aquatic, arboreal or rock-climbing reptiles. Should behavioural or breeding problems occur, or should further information on specific requirements for other species be required, advice should be sought from experts specialised in the species concerned and care staff, to ensure that any particular species' needs are adequately addressed. Additional information on species and habitats is available in the background information document by the expert group.

Where possible, reptiles used for experimental or other scientific purposes should be procured from reputable suppliers.

Table J.1.

Two habitat categories and examples of reptile species of each habitat frequently used

Habitat	Species	Size (cm)	Original geographic distribution/ Biotope	Optimal temperature	Relative humidity	Main period of activity
Aquatic	Trachemys scripta elegans Red-eared terrapin	20 to 28	Mississippi Valley drainage/Quiet water with muddy bottom	20 °C to 25 °C	80 to 100 %	Day
Terrestrial	Thamnophis sirta- lis Common gar- ter snake	40 to 70	North America/ Woodland, wet areas	22 °C to 27 °C	60 to 80 %	Day

# 2. The environment and its control

#### 2.1. Ventilation

Enclosures of reptiles should be adequately ventilated. To prevent animals from escaping, ventilation should be screen-covered.

## 2.2. Temperature

Reptiles are ectothermic. In order to maintain their body temperatures, under natural conditions they will select microenvironments in which they can gain or lose heat. Therefore, enclosures should offer to the animals areas of different temperatures (temperature gradient).

Temperature requirements of different species vary considerably and may even fluctuate in the same species at different times of the year. In the laboratory, room and water temperatures should be controlled. In many reptiles, sex determination and gonadal differentiation are temperature-dependent.

An incandescent lamp positioned over the platform provided as a resting board will allow basking reptiles to increase their body temperature. When the lights are turned off, a flat heating device may be used. Terraria of snakes or lizards from tropical biotopes should be furnished with at least one warmth-plate. Heating devices should be thermostatically-controlled to prevent the animals from overheating and burning.

## 2.3. Humidity

In order to regulate humidity, it will also be necessary to regulate the ventilation rate. A relative humidity of 70 to 90 % can be maintained by evaporating water from a container placed near the heater. The provision of areas of different humidity (humidity gradient) is beneficial.

# 2.4. Lighting

Appropriate light and dark regimes for each species, life stage, and time of the year should be provided. Reptiles should have the opportunity to withdraw to shaded areas within the enclosure. Light or sun lamps should not be the sole source of heat. The provision of ultraviolet radiation is necessary to stimulate the animal's production of vitamin D.

## 2.5. Noise

Reptiles are very sensitive to acoustic noise (airborne stimuli) and to vibratory noise (substrate-borne stimuli) and are disturbed by any new, unexpected stimulus. Therefore, such extraneous disturbances should be minimised.

# 2.6. Alarm systems

Adequate alarm systems should be provided if water circulation systems are used and/or aeration is required.

## 3. Health

Care is needed when housing different species of possible different health status.

## 4. Housing, enrichment and care

### 4.1. Housing

(See paragraph 4.5.2. of the General section)

#### 4.2. Enrichment

The habitat of reptiles should be structured to include, for example, natural or artificial branches, leaves, pieces of bark and stones. Reptiles benefit from such environmental enrichment in different ways: for example, such inclusions allow animals to hide, and provide labels for visual and spatial orientation. To prevent collision with clear glass, the side walls of the terraria should be patterned to provide a structured surface.

### 4.3. Enclosures — dimensions and flooring

Enclosures and enclosure furniture should have smooth surfaces and rounded edges to minimise the risk of injury, and in the most sensitive species opaque materials should be used.

### 4.3.1. Enclosures for aquatic reptiles

Aquatic reptiles should be accommodated in water-circulated, filtered, and aerated tanks. The water should be renewed about twice per week. To minimise the bacterial contamination of the water, water temperatures should not exceed 25 °C. Water levels should be sufficient for reptiles to submerge.

A platform should be provided as a resting board on which the reptiles can haul out or under which take shelter. Such platforms should be made of suitable materials, such as wood, so that animals are able to get a purchase with their claws in order to pull themselves out of the water. Platforms should be replaced at intervals as necessary. Platforms made of epoxy or polyurethane may not serve this function and will deteriorate quickly under continuous warm temperatures.

Table J.2.

Aquatic chelonians e.g., Trachemys spp.: Minimum enclosure dimensions and space allowances

Body length (*) (cm)	Minimum water surface area (cm²)	Minimum water surface area for each additional animal in group holding (cm²)	Minimum water depth (cm)
Up to 5	600	100	10
over 5 to 10	1 600	300	15
over 10 to 15	3 500	600	20
over 15 to 20	6 000	1 200	30
over 20 to 30	10 000	2 000	35
over 30	20 000	5 000	40

<sup>(\*)</sup> measured in a straight line from the front edge to the back edge of the shell

# 4.3.2. Enclosures for terrestrial reptiles

Terrestrial reptiles should be kept in enclosures consisting of an appropriate terrestrial part and an aquatic part. The water area of the terrarium should allow animals to submerge. It is advisable to renew the water at least twice a week, except in the case of a flow-through system.

Terraria should be transparent, have tight seams, with all holes securely screened, and be provided with well-fitted lids or doors that can be securely fastened down. All doors and lids should be fitted with latches, hooks or hasps. It is advisable to construct doors and lids, so that the entire top or an entire end or side opens to facilitate cleaning (except in the case of venomous reptiles). For some species, except for the front wall, all side walls including the top should be opaque. In case of highly irritable or easily frightened reptiles, the clear wall can be provided with a removable covering. For housing venomous snakes, certain security criteria must be fulfilled.

The provision of appropriate shelter is important for all terrestrial reptiles, both in which to hide and also sometimes to feed. A shelter-box, such as a tube of clay simulates the darkness of a burrow.

Table J.3.

Terrestrial snakes, e.g., Thamnophis spp: Minimum enclosure dimensions and space allowances

Body length (*) (cm)	Minimum floor area (cm²)	Minimum area for each additional animal in group-holding (cm²)	Minimum enclosure height (**) (cm)
Up to 30	300	150	10
over 30 to 40	400	200	12
over 40 to 50	600	300	15
over 50 to 75	1 200	600	20
over 75	2 500	1 200	28

<sup>(\*)</sup> measured from snout to tail

#### 4.4. Feeding

Captive reptiles should be maintained on their natural foods, foodstuffs or commercial diets approximating those of their natural diets. Many reptiles are carnivores (all snakes and crocodiles, most lizards, and some turtles), but some are vegetarian and others are omnivores. Some species exhibit very narrow and specific feeding habits. Reptiles, except for some snakes, can be trained to feed on dead prey. Therefore, it should normally not be necessary to feed on live vertebrates. When dead vertebrates are used, they should have been humanely killed using a method that avoids the risk of toxicity to the reptiles. Feeding regimes should be appropriate to the species, stage of development and husbandry system.

# 4.5. Watering

Drinking water should be provided for all reptiles.

## 4.6. Substrate, litter, bedding and nesting material

A variety of substrates may be used for terraria, depending on the requirements of the species. Fine sawdust and any other small-particle substrate should be avoided, as this may cause serious mouth or internal injuries or bowel obstruction, particularly in snakes.

### 4.7. Cleaning

(See paragraph 4.9. of the General section)

## 4.8. Handling

Care is needed when handling reptiles, as they can be easily injured. For example, some lizards may shed their tails (autotomy) if handled in an inappropriate way, and other species can easily be traumatised.

<sup>(\*\*)</sup> measured from the surface of the land division up to the inner part of the top of the terrarium; furthermore, the height of the enclosure should be adapted to the interior design including, e.g., shelves and large artificial branches

# 4.9. Humane killing

(See also paragraph 4.11. of the General section)

An appropriate method of killing is by an overdose of a suitable anaesthetic.

#### 4.10. Records

(See paragraph 4.12. of the General section)

#### 4.11. Identification

Where animals need to be identified individually a number of suitable methods are available: transponders; enclosure labels for individually housed animals; monitoring individual skin patterns (according to colour, skin damages, etc.); pen markings require renewal after skin shedding; small labels at the toes by coloured thread. Toe clipping is deleterious and should not be done.

## Transport

During transport reptiles should be provided with adequate air and moisture and, if necessary, appropriate devices included to maintain the required temperature and humidity.

#### K. SPECIES-SPECIFIC GUIDELINES FOR FISH

#### 1. Introduction

The use of fish as experimental animals has expanded greatly over the past decade for a number of reasons, including the great increase in aquaculture, which has led to a variety of supporting basic studies in areas such as nutrition, disease, physiology and genetics, ecotoxicology and other toxicological research, as well as fundamental studies in genetics and immunology whose results are of relevance to higher vertebrate groups, including mammals. A wide variety of fish species are used for experimental purposes and these have a diverse range of habitats, behaviour and environmental and husbandry requirements.

Fish are ectothermic animals and thus highly adapted to their particular aquatic environment. They react very rapidly to stress with immediate physiological consequences that can be relatively long-lasting and such changes, as well as having obvious welfare implications, will also impact upon experimental results.

Investigators and animal care staff should acquaint themselves with the characteristics of the proposed experimental fish species, to ensure that appropriate facilities and husbandry procedures are in place before animals are obtained. Species-specific guidance on rainbow trout (Oncorhynchus mykiss), Atlantic salmon (Salmo salar), tilapiine cichlids, zebra fish (Danio rerio), sea bass (Dicentrarchus labrax), Atlantic halibut (Hippoglossus hippoglossus), Atlantic cod (Gadus morhua), turbot (Scophthalmus maximus), African catfish (Clarias gariepenus) is available in the background document elaborated by the Group of Experts. Further advice on the requirements of these and other species should be sought from expert specialists and care staff to ensure that any particular species needs are adequately addressed.

During aquaculture research, when the aim of the research requires that fish are kept under similar conditions to those under which commercial fish are kept, the keeping of the animals should at least follow the standards laid down in the Directive 98/58/EC.

## 2. Environment and its control

## 2.1. Water supply

It is essential that an adequate water supply of suitable quality is provided at all times. Water flow in recirculatory systems or filtration within enclosures should be sufficient to remove suspended solids and wastes and to ensure that water quality parameters are maintained within acceptable levels. Monitoring systems should be in place to ensure fish are provided with an appropriate quantity of water of appropriate quality. Water flow should also be appropriate to enable fish to swim correctly and to maintain normal behaviour. In most cases, within enclosures housing post-larval fish, the water supply is best directed onto the water surface at an angle.

## 2.2. Water quality

Water quality is the most important factor in maintaining the well-being of fish and in reducing stress and the risk of disease. Water-quality parameters should at all times be within the acceptable range that sustains normal activity and physiology for a given species. The definition of acceptable range is complicated in that optimum conditions are not well defined for many species and that the requirements of individual species may vary between different life-stages e.g larvae, juveniles, adults or according to physiological status for example metamorphosis, spawning, feeding, previous history of exposure.

Fish show varying degrees of adaptability to changing water-quality conditions. Some degree of acclimatisation may be necessary and this should be carried out for a period appropriate for the fish species in question.

As most fish species cannot function well in water containing a high level of suspended solids, these should be maintained within an acceptable range. Where necessary water supply to facilities should be appropriately filtered to remove substances harmful to fish and to maintain suitable water physico-chemical parameters.

#### 2.2.1. Oxygen

Oxygen concentration should be appropriate to the species and the context in which they are held. Required oxygen concentration will vary according to temperature, carbon dioxide concentration, salinity, feeding level and amount of handling. Where necessary supplementary aeration of water should be provided.

## 2.2.2. Nitrogen compounds

Ammonia is the main excretory product of fish. Dissolved urea, as well as feed and faeces, are converted to inorganic compounds such as ammonia and phosphate. Ammonia will be further converted into nitrite and nitrate. Ammonia and nitrite are very toxic to fish and their accumulation should be avoided by increasing flow rate, reducing density or temperature, or biofiltration.

Susceptibility to ammonia varies between fish species and in general marine and younger fish are more susceptible. The toxic form of ammonia is unionised ammonia, the amount of which depends not only on total ammonia concentration, but also on pH, salinity and temperature.

## 2.2.3. Carbon dioxide (CO<sub>2</sub>)

Carbon dioxide is produced by fish during respiration and dissolves in water to form carbonic acid, thus lowering the pH. Accumulation of carbon dioxide can be a problem at a high stocking density if pure oxygen is used instead of air to maintain the oxygen content in the water. Although high concentrations of free carbon dioxide can be fatal to fish this is most unlikely to be a problem under normal housing conditions. However, care should be taken that water supply systems, particularly in the case of groundwater-based systems, do not introduce harmful quantities of carbon dioxide in the enclosures.

### 2.2.4. pH

Acceptable pH levels depend on many water quality factors, for example, carbon dioxide and calcium. As far as possible pH should be kept stable as any changes in pH will influence other water quality parameters. In general pH may be lower in freshwater than in salt water. If necessary supply water should be buffered.

# 2.2.5. Salinity

Salinity requirements of fish will vary according to whether they are marine or freshwater in origin or adapted. Some species are able to tolerate a wide range of salinity. In others salinity tolerance may vary according to life stage. Changes in salinity should be introduced gradually.

## 2.3. Temperature

Temperature should be maintained within the optimal range of the fish species involved and any changes should take place gradually. At high temperatures it may be necessary to provide supplementary aeration of enclosure water

## 2.4. Lighting

Many fish require light for feeding and other behavioural activities. Fish should be maintained on an appropriate photoperiod as far as possible since the day/night cycle influences the physiology and the behaviour of fish.

Many fish species should not normally be kept in bright light, although some tropical species naturally encounter very bright light. As appropriate for the species, lighting should be subdued or tanks should be covered and suitable hiding places provided. Abrupt changes in light should be avoided as far as possible.

#### 2.5. Noise

Fish can be acutely sensitive to sounds, even at very low levels. Noise levels within experimental facilities should be kept to a minimum. Where possible equipment causing noise or vibration, such as power generators or filtration systems, should be separated from fish-holding facilities. Fish reared in a particular environment will adapt to the stimuli presented there and may become stressed if moved to unfamiliar surroundings.

### 2.6. Alarm systems

(See Paragraph 2.6 of the General Section)

#### 3. Health

#### 3.1. General

Appropriate attention should be paid to hygiene within experimental facilities. The health of fish is intimately bound up with their environmental and husbandry conditions. Most diseases are associated with stress arising from deficiencies in these conditions and any attempt to control disease should address these areas if problems are to be successfully eradicated. Fish health management is almost always concerned with populations rather than single individuals, and control measures should be designed accordingly.

# 3.2. Hygiene and disinfection

Fish-holding facilities, including associated pipework, should be cleaned and disinfected when appropriate. In closed systems cleaning and disinfection should be compatible with maintenance of optimal microbiological conditions. Equipment, for example nets, should be disinfected between use. Staff should take precautions to prevent cross-contamination between fish enclosures.

## 3.3. Quarantine

Newly introduced stocks, both from farmed and wild fish, should be given an appropriate quarantine period, as far as possible separate from existing stocks. During quarantine they should be closely monitored and any disease problem which arises should be treated or the stock destroyed. Farmed fish should be procured from reputable suppliers and as far as possible have a verified health status.

# 4. Housing, enrichment and care

# 4.1. Housing

Fish behaviour will influence stocking density and schooling or territorial behaviour should be considered. The stocking density of fish should be based on the total needs of the fish in respect of environmental conditions, health and welfare. Fish should have sufficient water volume for normal swimming. Measures should be taken to avoid or minimise conspecific aggression without otherwise compromising animal welfare. Acceptable stocking density for a given species will vary depending on water flow and current, water quality, fish size, age, health and feeding method. In principle, groups should consist of fish of the same size to minimise the risk of injuries or cannibalism.

## 4.2. Enrichment

For some species, environmental enrichment may be necessary to take account of their behavioural traits, for example, in reproduction or predation. Examples of such needs include provision of hiding places for wrasse, or substrate such as sand for some flatfish. Care is needed to ensure that environmental enrichment does not

adversely affect water quality, but this should not impede the development of suitable measures to enhance the welfare of fish

#### 4.3. Enclosures

### 4.3.1. Fish holding facilities

Fish can be maintained in land-based enclosures in dedicated buildings or in external areas, or in enclosures in open-water systems. Where practical, these should have controlled access and be arranged to minimise disturbance of the fish, and to facilitate maintenance of suitable environmental conditions.

#### 4.3.2. Land-based enclosures

The materials used to construct the enclosures should be non-toxic, durable and with a smooth internal surface to prevent abrasions to the fish. Enclosures should be of an appropriate size to accommodate the required stocking density of fish and should be able to receive the necessary water flow. Enclosures should be of an appropriate shape to accommodate the behavioural needs and preferences of the particular experimental fish species; for example, circular enclosures are most appropriate for salmonids. Enclosures should be designed to prevent escape. Enclosures should where appropriate be self-cleaning to aid removal of waste products and surplus feed.

#### 4.3.3. Open-water enclosures

Fish, especially marine species, may be kept in large floating enclosures. The enclosure dimensions, including depth, should permit active swimming and shoaling of the fish. Mesh size should permit good water exchange while preventing escape of fish. Enclosures should be designed to minimise the risk of attack by predators. Enclosures should be rigged so as to prevent their shape distorting in tidal flows or running water and thus trapping fish.

## 4.4. Feeding

Fish may be fed either on artificial diet or fresh/frozen natural feed. Artificial diet is preferable, providing it meets the nutritional requirements of the species, and is acceptable to the fish. Some fish species or life stages will not take artificial diets. Artificial diets also tend to have less impact on water quality.

It is important that fish are fed at an appropriate feeding rate and frequency, and this will depend on a number of factors including temperature, size and maturity. As high temperature increases the metabolic rate, feeding level should also be increased. It may not always be necessary to feed fish daily. Presentation of diet is also very important to ensure adequate feeding. Consideration should be given to the number of meals per day, the age of the fish, the water temperature and the size of the pellet or food fragment offered. Feeding regime, palatability and the presentation of food should ensure that all fish obtain sufficient food. Particular attention should be paid to feeding of larval fish, especially where feeding is switched from live to artificial diets.

## 4.5. Cleaning of enclosures

All enclosures should be kept free of fish waste products or uneaten feed. If these are allowed to accumulate, water quality and thus fish health will be adversely affected. Enclosures should be regularly treated and cleaned to prevent fouling and reduced water exchange. There should be no risk of back-flushing and consequent fouling of enclosure water and the risk of infection. If enclosures are not self-cleaning, waste material should be siphoned off as necessary, generally as soon as possible after feeding. The sides and bottom of enclosures should be cleaned regularly to avoid build up of algae and other detritus. Care should be taken to minimise stress during cleaning.

### 4.6. Handling

Fish may be severely stressed by handling which should therefore be kept to the minimum possible. Fish should normally be netted out from the normal enclosure and anaesthetised in a smaller container before handling. Fish should be kept under anaesthetic for as short a time as possible and be placed in clean aerated water for recovery. An effective concentration of anaesthetic should be maintained throughout the procedure.

When catching fish, nets with an appropriate frame and mesh size should be used. Knotted net mesh should be avoided. Nets should be disinfected and rinsed in clean water before use.

Out of water fish should be handled with wet gloves or wet hands and on a moist surface to avoid scale and mucus loss. Particular attention should be paid to handling practices to avoid desiccation, suffocation and other injury.

## 4.7. Humane killing

Most fish should be killed by either:

- an overdose of anaesthetic using appropriate route and anaesthetic agent for the size and species. When killed by immersion, fish should be left in the anaesthetic solution for at least five minutes following the cessation of opercular movement and/or vestibulo-ocular reflex (VOR), or
- concussion of the brain by striking of the cranium

Death should be confirmed, for example, by physical destruction of the brain or exsanguination.

#### 4.8. Records

Records should be maintained on appropriate water quality parameters.

## 4.9. Identification

It is not always necessary or feasible to individually identify all fish within a facility.

If it is necessary to mark fish for identification purposes, subcutaneous dye injection is considered the least invasive method of marking. Careful consideration is needed before more invasive methods such as fin clipping or PIT tagging are used. Mechanical tagging should not be used unless no other method is suitable.

Marking should generally be carried out under anaesthesia in order to ease handling and minimise the risk of injury, morbidity and stress.

## Transport

Fish should be deprived of food prior to transportation for a period sufficient to allow the gut to clear and reduce faecal contamination of the transport system. Care should be taken to prevent injury and stress to fish during capture, loading, transportation and unloading. Abrupt temperature changes, periods of hypoxia and any deterioration in water quality due to excretory products should be avoided.